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GREENHOUSE CROPS PROTECTION AGAINST SUCTORIAL PHYTOPHAGES COMPLEX WITH THE PREPARATION BASED ON THE ENTOMOPATHOGENIC FUNGUS *LECANICILLIUM LECANII* (ZIMM.) ZARE & W. GAMS

Alena Yankouskaya, Dzmitry Voitka

Institute of Plant Protection, Belarus, Mira str. 2, 223011, ag. Priluki, Minsk region, e-mail: biocontrol@tut.by

Abstract

In production researches, the biological efficiency of preparation Entolek based on a strain *Lecanicillium lecanii* BL-2 is estimated. A significant inhibiting potential of the preparation for phytophages control in vegetable crops in greenhouses was determined: biological efficiency against the spider mite *Tetranychus urticae* Koch. was 94.6%, against onion thrips *Thrips tabaci* Lind. – 94.0%, against whitefly *Trialeurodes vaporariorum* Westw. – 90.7%. The application of the preparation Entolek in tomato crop provided the yield increase by 7.5-31.8%, in cucumber crop – 5.0-14.8%. The application regulations of the preparation Entolek for pest control in greenhouse crops, based on the pest number control and biological pest peculiarities, are developed.

Key words: biological control, bioinsecticide efficiency, *Lecanicillium lecanii, Tetranychus urticae, Trialeurodes vaporariorum, Trips tabaci.*

INTRODUCTION

While cultivating the greenhouse crops both in soil and subsoil and by small-volume hydroponics, a noxious damage is brought by a phytophages complex. Aphids, thrips, whitefly, spider mite are characterized by an increased harmfulness and a high reproductive potential, what necessitates the frequent crop treatments by pesticides, promotes

accumulation of their residues in a substrate and grown production (Akhatov et al., 2004). One of the dominant pests of the greenhouse crops is a common spider mite *Tetranychus urticae* Koch. – a polyphage damaging vegetable crops of fam. *Cucurbitaceae*, *Solanaceae*, *Leguminosae*, ornamental and flower crops. By feeding, a leaf mesophyll cells are damaged. At high colonization, the necroses, eventually covering the whole surface of a leaf, appear, the leaves are enlaced by a spider-web. The plant breath and photosynthesis are depressed, the yield is reduced. Greenhouse whitefly *Trialeurodes vaporariorum* Westw. spreads widely and makes a significant damage to cucumber, tomato, salad, celery crops and ornamental crops. As a noxious pest, it is noticed on 27 plant species, and damages 200 plant species among 82 botanical families. Whitefly larvae and adults, sucking sap from the plants leaves, strongly inhibit them. On the damaged leaves, the increased yellow spots gradually appear; after the hard damage, the leaves wither and die. The secretion of saprotrophic "sooty" fungi (*Cladosporium* sp.), colonizing on sugar pest, worsens the physiological plant state. Fungal bloom makes leaf assimilation of carbon dioxide difficult and leads to an overall plant suppression. The damaged leaves curl and dry up. The danger of this pest lies in its possibility to transfer certain viral diseases. Tobacco thrip *Thrips tabaci* Lind. is a polyphagous pest damaging different crops. In winter greenhouses where the pest reproduces parthenogenetically, up to eight generations of tobacco thrips may develop. By the pest imago and larvae feeding, the discolored areas appear on leaves which soon become of yellow and brown color. With the strong damage, the necrotic spots are produced, the leaves are deformed, dry out and break off easily. Tobacco thrips are the vectors of different viral diseases.

In this connection for the stabilization of phytosanitary situation, it is expedient to use ecologically friendly microbiological means of plant protection. An entomopathogenic fungus *Lecanicillium lecanii* (Zimm.) Zare & W. Gams makes a practical interest as a pathogen of arthopods. Literary sources identify an ability of *L. lecanii* to regulate the number of pest populations in protected ground (Chandler et al., 2005; Diaz et al., 2009; Patogeny..., 2001; Gurulingappa et al., 2011; Scorsetti et al., 2008; Vestergaard et al., 1995). The main advantage of this pathogen lies in the ability of a percutaneous penetration in

a host organism, and, hence, a suitability for an application for the control of suctorial pests (aphids, thrips, whitefly and spider mite), which aren't susceptible to contact and intestinal insecticides action. Demonstrating in natural conditions the properties of an obligatory parasite, *L. lecanii* preserves an ability to a saprotrophic type of nutrition and grows on artificial nutrition medium, what gives the opportunity to develop the preparations based on it (Devyatkina, 1985; Mitina et al., 2008).

Institute of Plant Protection (Belarus) has a collection of *L. lecanii* strains. At a preliminary stage of the researches, an insecticidal activity of the experimental samples of deep culture *L. lecanii* collection strains with respect to the main pests in the protected ground was studied while carrying out laboratory, vegetative and field trials. Screening results analysis of *L. lecanii* strains on productivity and insecticidal activity parameters let select *Lecanicillium lecanii* IZR BL-2 (Yankovskaya et al., 2011) strain as a preparation base. Estimation of the active mass spore material production of the strain *L. lecanii* BL-2 is done using a deep cultivation method, which is the most technological one for a large industrial production conditions of microbiological preparations. The research results are a base for the development of a new microbiological preparation Entolek.

The research aim is the development of the application technology of the preparation Entolek for the greenhouse crops protection against suctorial pests' complex.

The research objectives:

– To estimate the biological efficiency of a biopreparation on the phytophages number control;
– To work out the preparation application regulations.

METHODS

The researches were carried out in the *Open Joint-Stock Company* (OJST) "Greenhouse farm", *Joint-Stock Company* (JSC) "DorOrs" Minsk region in cucumber and tomato crops in conditions of small-volume cultivation technology in rock wool in winter-spring, spring-autumn and summer-autumn crop rotations in 2010-2012.

A biological preparation Entolek was produced according to TR BY 300042160.017-2011. The biological preparations Baciturin (on a base of *Bacillus thuringiensis* bacterium) and Pecilomicin-B (on a base of *Isaria fumosorosea* Wize (*Paecilomyces fumosoroseus*) entomopathogenic fungus) were used as a standard. The chemical preparations Actellic, EC (500 g/l pirimifos metil) and Fyfanon, EC (570 g/l malathion) were used as a chemical control in some experiments. The preparations were applied using the spraying method. A working solution rate was 600-1000 l/ha depending on a crop development stage.

Detection, identification and recording of pests were done according to common methods (Prischepa et al., 2008; Metodicheskie..., 2009). The pest number was recorded regularly (with 4-10 days interval) during vegetation period.

Estimation of yield data was done using samples selection method in 4 repetitions in every trial variant (Dospekhov, 1985).

The results of statistical analysis were processed using statistical analysis MS Excel packages (univariant dispersive analysis) considering the group differences between the average data based on less significant difference (LSD) with a probability of 95%.

RESULTS AND DISCUSSION

In our researches, a high sensibility of a spider mite *Tetranychus urticae* Koch. to the preparation Entolek is noticed in tomato crop. On the 4-7th days after treatment with the preparation, the pest death, caused by mycosis, is recorded: the damaged body of individuals became wizened and had a dark-brown colour. Bological efficiency of Entolek, after a single application, reached 59.6-94.6%, after two applications (with 7 days interval) – 83.0-86.0% (Zheronimo F_1, spring-autumn crop rotation). The biological efficiency after the third treatment of tomato plantings of summer-autumn rotation, carried out in connection with a pest number increase, grew from 59.5-67.2% to 91.0% (Cunero F_1, summer-autumn crop rotation) (Tables 1, 2). Thus, the preparation application let restrain the pest number from a critical increase without additional application of protective means during 1-2.5 months.

Table 1. *The effect of the preparation Entolek on the dynamics of a spider mite* Tetranychus urticae *Koch. number (OJST «Greenhouse farm», JSC «DorOrs», Minsk region, tomato Zheronimo F_1, spring-autumn crop rotation, small-volume technology of cultivation, rock wool, 2011)*

Variant	Mite average number, units per leaf / **Biological efficiency, %**									
	Before treatment	Day from the beginning of the trial								
		5th	11th	21st	29th	35th	60th	67th	75th	83rd
Entolek (5% w.s.)	0.3	0.03 **94.6**	0.03 **83.0**	0.36 -	0.3 -	0.2 **29.4**	0.9 -	0.13 **59.5**	0.1 **67.2**	0.53 -
Control group (without treatment)	0.56	1.03	0.33	0.63	0.33	0.53	2.7	0.6	0.57	0.47
LSD_{05}		0.94	0.25	0.3	0.1	0.4	1.2	0.37	0.43	0.09

Note. The repeated treatments were carried out on the 5th, and 64th days after the trial beginning.

Table 2. *The effect of the preparation Entolek on the dynamics of a spider mite* Tetranychus urticae *Koch. number (OJST «Greenhouse farm», JSC «DorOrs», Minsk region, tomato Cunero F_1, summer-autumn crop rotation, small-volume technology of cultivation, rock wool, 2011)*

Variant	Mite average number, units per leaf / **Biological efficiency, %**				
	Before treatment	Day from the beginning of the trial			
		6th	14th	21st	29th
Entolek (5% w.s.)	0.67	0.33 **59.6**	0.2 **86.0**	0.23 -	0.17 **91.0**
Control group (without treatment)	0.33	0.4	0.7	0.1	0.93
LSD_{05}		0.09	0.37	0.2	0.56

Note. The repeated treatments were carried out on the 6th, 21st days after the trial beginning.

The preparation application let obtain the yield increase during tomato harvesting from 7.5% (at the end of vegetation) to 14.3% (during mass fruiting).

Table 3. *The effect of the preparation Entolek on the dynamics of a spider mite* Tetranychus urticae *Koch. number (OJST «Greenhouse farm», JSC «DorOrs», Minsk region, cucumber Courage* F_1*, summer-autumn crop rotation, small-volume technology of cultivation, rock wool, 2011)*

Variant	Mite average number, units per leaf / **Biological efficiency, %**			
	Before treatment	Day from the beginning of the trial		
		8th	15th	21st
Entolek (5% w.s.)	0.8	1.65 **10.7**	0.4 **70.2**	1.97 -
Chemical control group	1.1	2.55	1.85	2.53
LSD_{05}		1.2	1.7	0.87

Note. The repeated treatment was carried out on the 11th day after the beginning of the trial.

Table 4. *The effect of the preparation Entolek on the dynamics of a spider mite* Tetranychus urticae *Koch. number (OJST «Greenhouse farm», JSC «DorOrs», Minsk region, cucumber Courage* F_1*, winter-spring crop rotation, small-volume technology of cultivation, rock wool, 2012)*

Trial variant	Mite average number, units per leaf / **Biological efficiency, %**						
	Before treatment	Day of the record					
		7th	14th	21st	34th	42nd	49th
Entolek (5% w.s.)	0.47	0.34 **59**	0.80 **51.8**	1.14 **79.1**	2.94 **83,2**	1,47 **93,3**	2,07 **75**
Baciturin (1% w.s.)	0.80	0.60 **57.5**	7.07 -	1.40 **85**	2.47 **91.7**	3.47 **88.1**	2.40 **83**
Control group (without treatment)	0.34	0.60	1.2	3.94	12.67	12.34	6.00
LSD_{05}		0.16	0.2	2.1	4.4	7.31	2.91

Note. The repeated treatments were carried out on the 7th, 14th, 34th, 42nd days

While applying Entolek against a spider mite in cucumber crop, the preparation efficiency was also high. The application scheme of the preparation for the cucumber protection against the phytophage

included 2 times application of 5% working solution by spraying from a moment of detection of the first pest individuals with 7-10 days interval. Subsequently, the preparation was applied (up to 5 times) based on phytosanitary monitoring data during the spider number increase. The protective measures served as a control group were carried out by the greenhouse farm (insecticidal treatments: Aktellik, EC; Fyfanon, EC). Efficiency of a single treatment on the 7th day increased from 10.7 to 59.0%, of double treatment (with the 7-10 days interval) – from 51.8 to 70.2% (Tables 3, 4).

The application of the preparation Entolek promoted a stable increase of the yield at total cucumber harvesting in 5.0-14.8%.

While carrying out the researches in cucumber crop of winter-spring crop rotation, a comparison between the efficiency of Entolek and the bacterial preparation Baciturin (1% w.s., 12-30 l/ha) was done. The treatments with preparations were carried out from the moment the first spider mite individuals appeared on the crops (planting out on a permanent place in a greenhouse). Efficiency of the Entolek treatments was high (51.8-93.3%) and exceeded a variant with the Baciturin application (57.5-91.7%) (Table 5). Maximal efficiency was noticed on the 42nd day of the trial – 93.3% after the third time of the preparation application. The protective action of a preparation has lasted for 7-14 days. Protective effect of Entolek let obtain 5.0-14.8% more cucumber fruits during mass fruiting (Table 3), while application of Baciturin – only 4.3% more.

Efficiency of application of Entolek against a spider mite was higher and protective action periods were longer while applying it on tomato crop. This can be related to the fact that tomato is less preferred plant by the pest in comparison with the cucumber, what increases the phytophage susceptibility to external factors, including the biopreparations (Akhatov et al., 2004). As a rule, later colonization of tomato plantings with a spider mite is determined by this fact, therefore, Entolek was predominantly applied in the middle of the vegetation (mass fruiting period), while in the cucumber crop, often in the beginning (planting out on a permanent place – beginning of fruiting).

In order to evaluate the effectiveness of a biological preparation with respect to the greenhouse whitefly *Trialeurodes vaporariorum* Westw., the preparation Entolek was used 2-3 times in the form of plants

spraying as a 5% working solution. The preparation Pecilomicin–B (1% w.s., 40 kg/ha) following the same scheme was used as a variant for the comparison, as a control group – the protective measures carried out by the farm (single treatment by the preparation Fyfanon, EC). The effective single treatment with Entolek has reached 57.1-78.0% effectiveness, Pecilomicin-B – 67.2%, twofold treatment – 71.4-90.7% and 88.3%, accordingly, indicating a similar level of biological activity of preparations (Tables 5, 6).

Table 5. *Biological efficiency of the biological preparation Entolek for the control of whitefly* Trialeurodes vaporariorum *Westw. number (field trial, OJST «Greenhouse farm», JSC «DorOrs», tomato ZheronimoF$_1$, summer-autumn crop rotation, small-volume technology of growing, rock wool, 2011)*

Experiment variant	Average pest number, units per leaf / **Biological efficiency**, %					
	Before treatment	Day of the record				
		6th	13th	19th	27th	34th
Entolek, l. (5%- w.s.)	0	0.03 **57.1**	0.6	0.1 **80.0**	0.2 **71.4**	0.06 **53.8**
Control group (without treatment)	0.03	0.07	0.13	0.5	0.7	0.13
LSD_{05}		0.05	0.52	0.32	0.29	0.07

Note. The repeated treatments were performed on the 6th and 19th day.

Table 6. *Biological efficiency of the biological preparation Entolek for the control of whitefly* Trialeurodes vaporariorum *Westw. number (field trial, OJST «Greenhouse farm», JSC «DorOrs», tomato Zheronimo F$_1$, summer-autumn crop rotation, small-volume technology of growing, rock wool, 2012)*

Experiment variant	Average pest number, units per leaf / **Biological efficiency, %**			
	Before treatment	Day of the record		
		7th	17th	24th
Entolek (5%- w.s.)	0.67	0.13 **88.0**	0.33 **78.0**	0.14 **90.7**
Pecilomicin-B (1% w.s.)	0.53	0.73 **19.8**	0.4 **67.2**	0.13 **88.3**
Chemical control	0.47	0.8	1.07	1
LSD_{05}		0.52	0.74	0.79

Note. The repeated treatment was done on the 17th day.

It is noted that the increase in rate of pest population number during researches was lower than that of common spider mite, what explains the necessity of fewer treatments and longer time interval between treatments. By reducing the phytophage number, 11.0-38.4% of tomato yield has been saved.

While carrying out the researches on cucumber crop Cerez F_1 and Courage F_1, an essential inhibitory effect of Entolek on tobacco thrip *T. tabaci* is observed. This pest, in addition to high fecundity and harmfulness, is characterized by an active and fast settling when released into a greenhouse and, consequently, the increased risk of mass reproduction. However, the evaluation of the influence of Entolek application on the pest population dynamics pointed out its significant effect on population decrease, on the average, from 2.05 to 0.1 individual per leaf during 6 weeks. According to the evaluation results, the biological efficiency of twofold treatment with the biological preparation with 7 days interval has reached 90.0-94.0% for 3 weeks (Courage F_1, summer-autumn rotation) (Table 7).

Table 7. *The influence of the preparation Entolek on tobacco thrips* Thrips tabaci *Lind. number (OJST «Greenhouse farm», JSC «DorOrs», Minsk region, cucumber Courage F_1, summer-autumn crop rotation, small-volume cultivation, rock wool, 2010)*

Variant	Average number of tobacco thrips, units per leaf / **Biological efficiency, %**			
	Before treatment	Day from the beginning of the trial		
		8th	15th	41th
Entolek (5% w.s.)	2.05	0.6 **10.0**	0.2 **94.0**	0.1 **90.0**
Control (without treatment)	0.3	0.1	0.5	0.15
LSD_{05}		0.58	0.22	0.07

Note. The repeated treatment was done on the 11th day from the beginning of the trial.

CONCLUSIONS

The results of researches have shown that the biological preparation Entolek, based on a high-active entomopathogenic fungus strain *Lecanicillium lecanii* BL-2, renders a significant inhibitory potential in the control of the greenhouse suctorial phytophages – common spider mite *Tetranychus urticae* Koch., tobacco thrips *Thrips tabaci* Lind., greenhouse whitefly *Trialeurodes vaporariorum* West.

The biological efficiency of the preparation in relation to the spider mite has reached 51.8-93.35%, tobacco thrips – 90.0-94.0%, and greenhouse whitefly – 57.1-90.7%. The application of the preparation Entolek promoted tomato crop yield increase by 7.5-31.8%, cucumber – by 5.0-14.8%.

The results of the analysis of carried out experiments let recommend the biological preparation Entolek as means of control of biological greenhouse crop pest and the following common scheme of application is proposed: initiate the application at the appearance of the first phytophage local focuses, and carry on with a twofold treatment with 5% w.s. of a preparation with the 5-10 days interval. The subsequent treatments are needed in case the pest population number increases, with the 5-7 days interval for the most aggressive species (spider mite, tobacco thrips) and 7-14 days – for the greenhouse whitefly.

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Alena Yankouskaya, Dzmitry Voitka

ŠILTNAMIUOSE AUGINAMŲ AUGALŲ APSAUGA NUO ČIULPIANČIŲJŲ FITOFAGŲ KOMPLEKSO PANAUDOJANT PREPARATĄ, GAUTĄ IŠ ENTOMOFAGINIO GRYBO *LECANICILLIUM LECANII* (ZIMM.) ZARE & W. GAMS

Santrauka

Buvo atlikti preparato *Entolek*, kurio pagrindas *Lecanicillium lecanii* štamas BL-2, efektyvumo tyrimai. Nustatytas žymus preperato slopinantis poveikis fitofagams, kenkiantiems daržovėms šiltnamiuose: biologinis aktyvumas prieš voratinklinę erkę (*Tetranychus urticae* Koch.) – 94,6 %; prieš svogūninį tripsą (*Thrips tabaci* Lind.) – 94,0 %; baltasparnį (*Trialeurodes vaporariorum* Westw.) – 90,7 %. Preperato *Entolek* panaudojimas pomidorų derlių padidino 7,5–31,8 %, agurkų – 5,0–14,8 %. Preparato *Entolek* veikimas šiltnamiuose auginamoms daržovėms apsaugoti pasireiškia kenkėjų skaičiaus sumažinimu ir poveikiu kenkėjų biologijai.

Reikšminiai žodžiai: biologinė kontrolė, biologinių insekticidų efektyvumas, *Lecanicillium lecanii*, *Tetranychus urticae*, *Trialeurodes vaporariorum*, *Thrips tabaci*.