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## Use of biological and chemical fungicides and biostimulators for potato protection against early and late blights

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**Summary:** Ecological risks associated with chemical pesticides encourage application of biopesticides and integrated pest management. The study compared efficiency of the Manfil fungicide and Agat-25K biopreparation (single or combined with the ExelGrow biostimulator) against the late (LB) and early (EB) blights of potato. Field experiments were conducted with three potato cultivars (Arizona, Vektor, Aluett) of different resistance levels to the LB and EB diseases. Leaf blight development was registered as LB (Arizona), LB+EB (Vektor), and EB (Aluett). Agat-25K delayed LB development under epiphytotic conditions on the susceptible cv. Arizona (biological efficiency 39%), though was inferior to Manfil (62%). On the contrary, for cv. Aluett, Agat-25K showed more significant EB suppression than Manfil (96 vs. 83%). Compared to the control, the yield increase of cv. Arizona was 15.8 t/ha for Agat-25K and 25.4 t/ha for Manfil. Tuber marketability increased by 28 and 13% for Agat-25K and Manfil, respectively. For moderately resistant and resistant cultivars (Vektor, Aluett), Agat-25K provided the highest yield increase compared to Manfil (19.1 vs. 16.7 t/ha for Vektor and 4.5 vs. 1.4 t/ha for Aluett, respectively), while the tuber marketability increased by 15% (Vektor) and 18% (Aluett). Agat-25K can be recommended for use on moderately-resistant cultivars under sporadic LB epiphytoses and for the EB control. ExelGrow application with Manfil and Agat-25K was first shown to improve their biological efficiency on moderately LB-resistant and LB-resistant cultivars (up to 12%).

**Keywords:** *Alternaria solani*, biopreparations, integrated crop protection, leaf blights, *Phytophthora infestans*, potato

### Introduction

Being the third largest staple food crop in a global scale, potato is very important for the food safety of many countries (Beumer et al., 2021). However, the productivity and yield of this crop are significantly influenced by a number of plant pathogens. The most important diseases of potato associated with the vast majority of disease management costs and activity in the temperate zones are leaf blights (late and early blights). Both diseases also affect tuber quality and storability. Early blight

(EB) of potato, caused by *Alternaria solani* Sorauer can result in up to 30% annual yield losses (Leiminger et al., 2014). Late blight (LB) of potato caused by the oomycete *Phytophthora infestans* (Mont.) de Bary imposes an additional “tax” on potato growers, which may reach up to \$3–10 billion per year including direct yield losses and management costs (Dong and Zhou, 2022). According to our data, in recent years LB is manifested unusually early (shoot appearance phase) in some regions of Russia with a rapid development. An increased epidemiologic potential of *P. infestans* causes a significant loss in efficiency of common potato protection activities.

The most common and effective way to control fungal crop diseases in the case of an intensive agriculture is the use of chemical pesticides. However, due to the pathogens’ ability to develop fungicide-resistant forms, plant growers have to either increase the frequency or dosages of fungicides used (extensive way), or use integrated systems of crop protection, which include treatments with a number of fungicides characterized by different modes of action. This results in the increased soil pollution with pesticide residues thereby increasing environmental and health risks. Due to health concerns arising from the pesticide risk assessment regularly performed by the experts of the European Food Safety Authority (Markantonis et al., 2018; Chatzidimitriou et al., 2019), and the opinion of consumers, who consider pesticides as a threat, the maximum residue levels of commercial pesticides are regularly revised towards their reduction. Moreover, in the next decade, the range of authorized plant protection products in Europe is expected to decrease due to banning or restricting their use (Erokhova and Kuznetsova, 2022; Kim et al., 2017). Thus, many authorities encourage the use of biopesticides as a part of the integrated pest control for providing environmental safety (Ong et al., 2020).

The aim of this study was a comparative evaluation of the biological and economical efficiency of a commercially available fungicide Manfil and a complex Agat-25K biopreparation application (both either single or combined with the ExelGrow biostimulator) on three potato cultivars with different levels of resistance to the late and early blights of potato.

## Materials and Methods

*Potato cultivars and pesticides used in the study.* Three potato cultivars differing in their LB and EB resistance were used in the study: Arizona (susceptible to LB, moderately susceptible to EB), Vektor (moderately resistant to LB and EB), and Aluett (resistant to LB, susceptible to EB).

Manfil 80 WP (a.i. mancozeb, 800 g/kg, Indofil Industries Ltd., Mumbai, India), a commercial contact fungicide intended to protect potato against LB and EB, was used as a chemical fungicide. Agat-25K (Edna Ltd., Moscow, Russia), is a commercial biopreparation based on metabolites of *Pseudomonas aureofaciens* strain H16 (VKM B-2433 D) and intended to protect potato against LB and EB. The preparation also includes a complex of growth stimulators, such as indole-3-acetic acid (IAA),  $\alpha$ -alanine,  $\alpha$ -glutamic acid, as well as balanced mix of micro- and macroelements, soybean seedling autolysate, pine needle extract, and chlorophyll-carotene pine needle paste (Kuznetsova, 2000). ExelGrow preparation (Adama Russia, Moscow, Russia) based on the *Ascophyllum nodosum* alga was used as a biostimulator.

Protective treatments of potato cultivars with chemical and/or biological preparations were arranged in accordance with the routine scheme (six treatments performed each 7–10 days); the first treatment was carried out on June 29, 2023. Application of the ExelGrow biostimulant included the first treatment at the bud formation phase followed by two consecutive treatments performed 7 and 14 days later.

*Land treatment and design of the experiment.* The study was arranged in 2023 at the experimental field of the All-Russian Research Institute of Phytopathology (ARRIP, Moscow region, Russia). The land treatment corresponded to the common practice used at ARRIP for potato production and included under-winter ploughing, disking, deep ground treatment, pre-planting furrow formation, and hilling. Organic (70 ton/ha) and inorganic (60 kg/ha) fertilizers were applied in autumn and spring (prior potato planting), respectively. Herbicidal treatments included the use of Zencor (a.i. metribuzin, 0.6 kg/ha, Bayer AG, Leverkusen, Germany) and Boxer (a.i. prosulfocarb, 2 L/ha, Syngenta, Basel, Switzerland) at the end of May and June, respectively. During a vegetation season, the whole field was once treated with a thiamethoxam-based Aktara insecticide (0.06 kg/hectare).

*Planting.* Potato cultivars were planted on May 5, 2023 and manually harvested on Aug 20, 2023. The area of each experimental plots was 42 m<sup>2</sup>, and the plots were randomly distributed across the field part planted with the corresponding cultivar. The experimental design included five different variants of protection, each in four replications (Table 1).

*Registration of the LB and EB development and data analysis.* To evaluate the LB and EB development, regular field inspections were carried out between June 29 and August 15. The level of the disease development was evaluated according to the British Mycological Society scale (James et al., 1972); the corresponding AUDPC values were calculated according to Shaner and Finney (1977). The crop yield was determined right after a manual harvesting of plots. The statistical analysis of the obtained data was carried out by ANOVA at the 95% confidence level (Dospikhov, 1984). All data (except dynamics of the leaf blight development) are shown with the corresponding least significant value (LSD<sub>0.95</sub>) values. Biological efficiency of the tested protection schemes was evaluated using the Abbott's formula (Abbott, 1925).

Table 1. The scheme of plant protective treatments

Variant no.	1	2	3	4	5 (Control)
1 <sup>st</sup> treatment (29.06.2023)	Manfil	Manfil	Agat-25	Agat-25	–
2 <sup>nd</sup> treatment (05.07.2023)	Manfil	Manfil + ExelGrow	Agat-25	Agat-25+ Exelgrow	–
3 <sup>rd</sup> treatment (12.07.2023)	Manfil	Manfil + ExelGrow	Agat-25	Agat-25+ Exelgrow	–
4 <sup>th</sup> treatment (20.07.2023)	Manfil	Manfil + ExelGrow	Agat-25	Agat-25+ Exelgrow	–
5 <sup>th</sup> treatment (27.07.2023)	Manfil	Manfil	Agat-25	Agat-25	–
6 <sup>th</sup> treatment (07.08.2023)	Manfil	Manfil	Agat-25	Agat-25	–

*Note.* Preparation doses used for the treatment: Manfil, 1.6 kg/ha; Agat-25, 140 g/ha; ExelGrow, 1 L/ha.

## Results and Discussion

*Effect of the tested treatments on the development of leaf blights in potato.* Weather conditions of 2023 characterized by temperature and humidity drops in the beginning of the vegetation season and abundant rainfalls in June and July were favorable for the early and active LB development. This resulted in the early LB manifestation on the LB-susceptible cv. Arizona (June 29 for the control variant). In the first and third decades of July, the level of infection of the control plants was 20 and 85%, respectively; to the first decade of August, this index reached 100% (Fig. 1, top). Due to this fact, the dynamics of the leaf blight development on this cultivar was provided by the LB only.

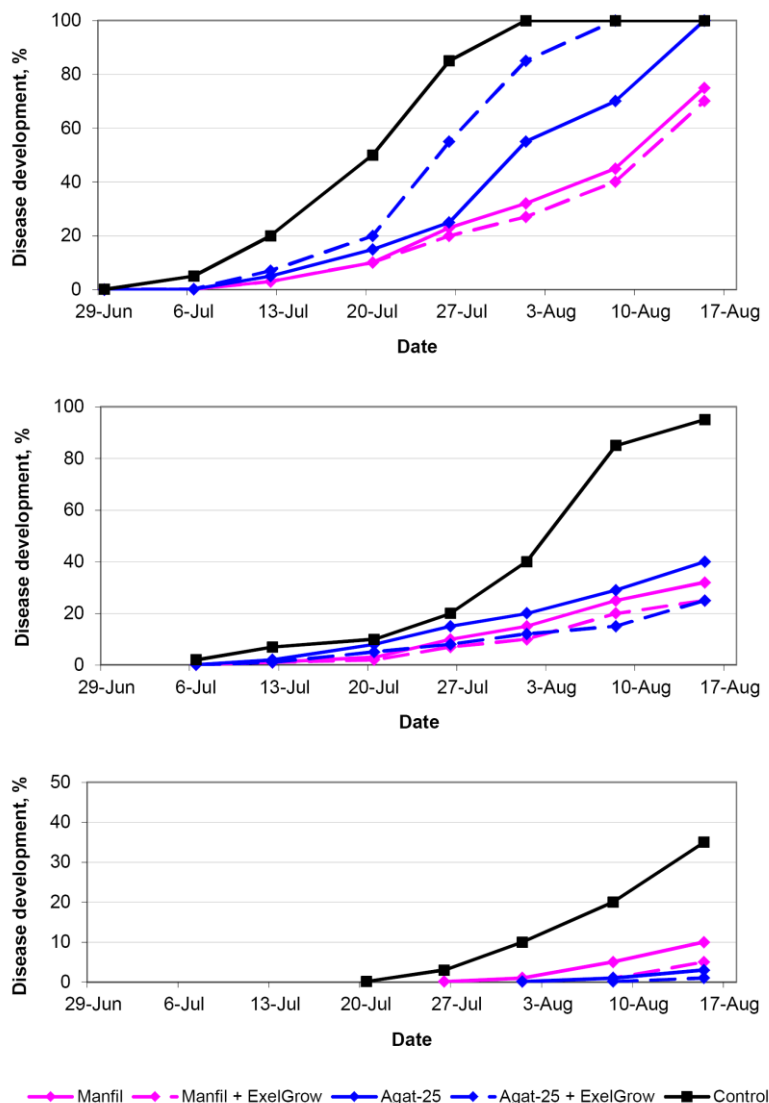


Figure 1. Dynamics of the leaf blight development in experimental treatments recorded for three potato cultivars: Arizona (top), Vektor (middle), and Aluett (bottom).

For the cv. Vektor characterized by a moderate resistance to both LB and EB, the first LB manifestations in the control variant were observed on July 4 (Fig. 1, middle). To the end of July, the disease development reached 20% and significantly increased to the Aug 15 (95%). The main contribution to the plant affection with the leaf blight was made by LB, since the EB development started significantly later.

In the case of the LB-resistant, but EB-susceptible cv. Aluett, no LB infection on leaves was registered for the whole observation period, while an active EB development was observed. The first EB manifestations in the control variant were registered on July 20 (Fig. 1, bottom). Thus, EB was the only contributor to the observed leaf blight infection of this cultivar.

The analysis of the obtained results showed that the application of the Manfil fungicide not only delayed the first LB appearance on the susceptible cv. Arizona, but also restrained its further development. Use of the mix of Manfil and ExelGrow preparations did not provide any significant impact on the disease development compared to the use of Manfil alone, though increased the yield

(see the next subsection). The efficiency of the Agat-25K biopreparation was inferior to that of Manfil. Moreover, application of Agat-25K in the mix with the ExelGrow biostimulator was less efficient than Agat-25K alone. According to the AUDPC calculations, the best biological efficiency for this cultivar was obtained in the variants Manfil and Manfil+ExelGrow (62 and 66%, respectively, Table 2).

Table 2. AUDPC values and biological efficiency of the treatments for leaf blights control on different potato cultivars

Treatment	cv. Arizona		cv. Vektor		cv. Aluett	
	AUDPC, rel. units	Biological efficiency, %	AUDPC, rel. units	Biological efficiency, %	AUDPC, rel. units	Biological efficiency, %
Manfil	1015	62	461	70	62	83
Manfil + ExelGrow	912	66	346	78	19	95
Agat-25K	1625	39	619	60	15	96
Agat-25K + ExelGrow	2127	21	332	78	2	99
5 (Control)	2685	–	1540	–	363	–
LCD <sub>0.95</sub> *	130		40		8	

\* The least significant difference calculated by ANOVA.

In the case of the moderately LB- and EB-resistant cv. Vektor, we observed the development of both LB and EB diseases. Crop treatments with Manfil or Agat-25K preparations provided more late manifestation of leaf blights and the further control of their development. Though being inferior to Manfil in efficiency, Agat-25K still significantly delayed the development of both diseases for the whole vegetation season compared to the control. Combination of each of the preparations with the ExelGrow biostimulator improved their protective efficiency and provided some yield increase (see the next subsection). Calculations of AUDPC values showed that the best biological efficiency for this cultivar was obtained in the Manfil+ExelGrow and Agat-25K+ExelGrow variants (78% for each variant, Table 2).

As it was mentioned earlier, the LB-resistant and EB-susceptible cv. Aluett showed the first EB manifestations in the control variant on July 20. For the variant protected by Manfil, the corresponding date was August 1, whereas application of Agat-25K shifted this date to August 8. Combination of both preparations with the ExelGrow biostimulator provided a significant delay in the disease manifestation compared to their single application (6 and 7 days for Manfil and Agat-25K, respectively). Therefore, Agat-25K provided the best level of potato protection against EB (96% of biological efficiency), and the ExelGrow addition to the protection schemes resulted in an additional improvement of their efficiency (from 83 to 95% for Manfil and from 95 to 99% for Agat-25K).

*Effect of the tested plant protection treatments on the yield and marketability of potatoes.* The data for the yield and marketability of harvested tubers are shown in Fig. 2. For the cv. Arizona, the maximum additional yield (compared to the control) was registered for the Manfil and Manfil+ExelGrow variants (92.7 and 95.62%, respectively); the corresponding increase in the tuber marketability was 28 and 29%, respectively (Fig. 2, top). Less efficient protection revealed for Agat-25K also influenced on the yield and its quality. Compared to the control, the yield increase for the Agat-25K and Agat-25K+ExelGrow variants was 57.7 and 22.3%, respectively, and the marketable fraction of tubers increased by only 13 and 3%, respectively.

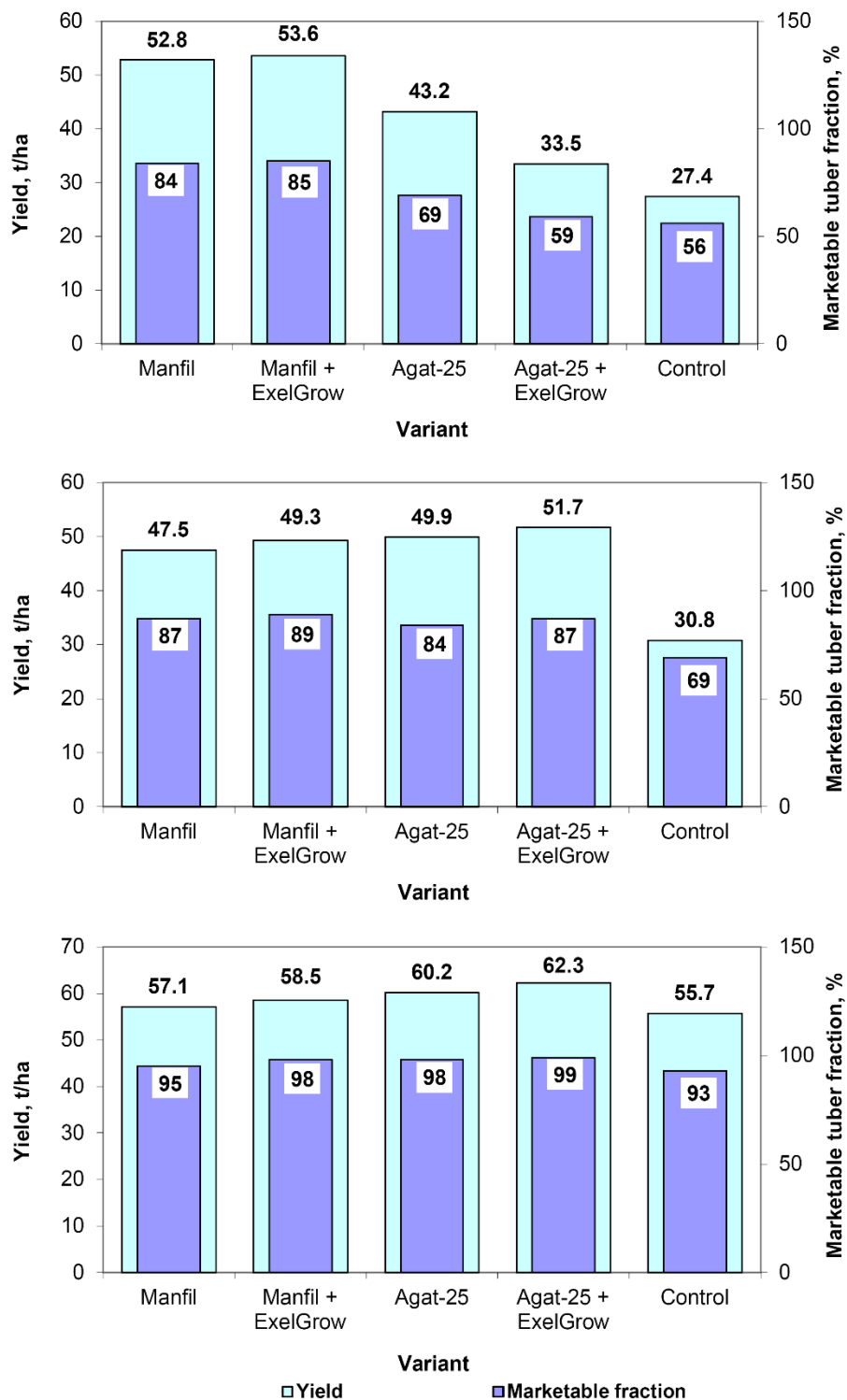


Figure 2. Yield and marketability of potatoes in the tested plant protection treatments determined for three potato cultivars: Arizona (top), Vektor (middle), and Aluett (bottom). The least significant difference ( $LSD_{0.95}$ ) values for the yield are 30 (cv. Arizona), 18 (cv. Vektor), and 20 (cv. Aluett). For the fraction of marketable tubers,  $LSD_{0.95} = 3$  for all three cultivars.



Thus, for the LB-susceptible cultivar, the results (biological efficiency of the LB protection, yield, and tuber marketability) obtained for the Manfil fungicide demonstrate its high protective efficiency under conditions of the epiphytotic LB development. The Agat-25K biopreparation also delayed the LB development, though was inferior to the chemical fungicide. Use of the ExelGrow biostimulator together with the Manfil fungicide did not significantly improve the protective efficiency of this fungicide, though provided some yield increase (+0.8 t/ha); at the same time, its addition to the Agat-25K biofungicide worsened the results compared to the Agat-25K only.

In the case of the moderately resistant cv. Vektor, a significant increase in the yield (54.2–67.86%) and marketable tuber fraction (15–20%) was observed for all tested schemes of protection (Fig. 2, middle). Therefore, both Manfil and Agat-25K provided high protective efficiency under conditions of the LB epiphytotic and active EB development. The best results were obtained for Agat-25K: in this case, the yield increase significantly exceeded that observed for the Manfil application either for the single fungicide application, or in combination with ExelGrow (62.0 and 67.9% vs. 54.2 and 60.1%, respectively).

As for LB-resistant and EB-susceptible cv. Aluett, application of the Manfil fungicide insignificantly increased the yield compared to the control (1.4 t/ha or 2.5%), whereas Agat-25K provided a significant increase (4.5 t/ha or 8.1%). Combination of these preparations with ExelGrow provided the yield increase by 5.0 and 11.9% compared to the control. Since the marketable fraction of tubers in the control was high (93%), the increase of this index provided by the tested protection schemes was not too high (2–6%), though significant in all cases excepting the single Manfil application; no significant difference in this index was observed between Manfil+ExelGrow, Agat-25K, and Agat-25K+ ExelGrow variants.

The observed yield increase (8.1–57%) in potato cultivars treated with Agat-25K is in line with the data of earlier studies, in which application of the Agat-25K on various potato cultivars resulted in the average yield increase varied from 10–12% (Statsyuk et al., 2015; Semenchuk, 2018) to 20–30% (Shlyakhov and Grigoryan, 2017; Uromova et al., 2016). Such increase can be explained by several reasons. The first reason is the growth-regulating activity of this biopreparation confirmed on a number of crops, such as soybeans (Romanova and Gins, 2006), earth apple (Plyasunov et al., 2022), sunflower (Avdeenko et al., 2021), potato (Uromova et al., 2016; Semenchuk, 2018), etc. Acceleration of the crop development by Agat-25K may result in the earlier passing of plants through growth stages vulnerable for pathogen infections and pest attacks, and/or periods characterized by an increased risk of infection thus reducing the disease development and improving the yield (Kuznetsova, 2000; Sadykov et al., 2016). Another reason is the announced ability of Agat-25K to elicit plant immunity thus improving its resistance to a range of diseases capable of reducing the tuber yield (Romanova and Gins, 2006; Uromova et al., 2016). Finally, some contribution to the yield increase was made by the direct antifungal effect of Agat-25K. This effect was confirmed in a number of publications (Marijanovski et al. (2015) who reported that Agat-25K provided a strong suppression of some fungal diseases of tomato and pepper crops, such as *Phytophthora infestans*, *P. capsici*, *Alternaria solani*, *Botrytis cinerea*, etc. Pre-sowing treatment of barley and wheat with this biopreparation reduced the development of Septoria and root rot infections of plants by 40 and 56–64%, respectively (Marinchenko, 2020). Nikolaeva and Lukina (2022) reported that the treatment with Agat-25K significantly reduced potato susceptibility to diseases (from 19 to 6–7%). Uromova et al. (2016) reported that the Agat-25K treatment provided a 3-fold reduction of the occurrence and development rate of the late blight on two potato cultivars – Udacha and Red Scarlett. In our study, the revealed AUDPC reduction provided by this biopreparation varied from ~40 to 96% proportionally to the level of the cultivar resistance. The existing antifungal activity of Agat-25K should provide a greater contribution to the yield formation and quality of tubers in susceptible cultivars that was confirmed

by our results (57% yield increase in LB-susceptible cv. Arizona, 22.8% yield increase in moderately LB-resistant cv. Vektor, and 8.1% yield increase in LB-resistant cv. Aluett).

The ExelGrow stimulator was recently shown to provide a slight yield increase (~7%) when used alone to protect potato against EB; being combined with some chemical fungicides (Miravis, Revus Top, Signum), it improved the same indices for about the same degree (7.7–8.1%) (Ukolova et al., 2024). At the same time, its combination with these fungicides was first shown to provide a significant decrease in the EB infection of potato (18, 11, and 12%, respectively) compared with the same fungicides applied alone; in terms of marketability, the average yield improvement provided by this biostimulator made 20–22%. These results, as well as those obtained in this study, indicated good prospects for the use of ExelGrow to improve the protecting efficiency of biological and chemical fungicides against EB. To the author's best knowledge, in these two studies, the influence of this biostimulator combined with fungicide preparation on the suppression of plant diseases was evaluated for the first time.

Taking into account the obtained results, one can recommend to use Agat-25K in regions characterized by sporadic LB epiphytoses as well as on moderately resistant cultivars. Also, Agat-25K provided quite good protective efficiency under conditions of the active EB development, and can be recommended for use to control this disease on susceptible cultivars. Application of the ExelGrow biostimulator improved the protecting efficiency of both preparations against leaf blights and provided a significant yield increase (+1.8 t/ha for both cases), though rather small increase in the marketable tuber fraction.

## Conclusions

The performed study provided new experimental data on the biological and economical efficiency of the tested plant protection treatments of potato cultivars differing in their LB and EB resistance. The Agat-25K biological preparation was shown to delay the LB development under epiphytotic conditions on susceptible cv. Arizona, though it was inferior to the contact chemical fungicide (Manfil). However, in the case of moderately resistant and resistant cultivars (Vektor and Aluett), use of the Agat-25K biopreparation provided better yield compared to the Manfil application; in the case of cv. Aluett, it also showed a more significant EB suppression. Obviously, Agat-25K possesses a dual action: it decreases the leaf blight severity and activates plant growth and development. Taking into account this fact, Agat-25K can be recommended for use on moderately resistant cultivars under conditions of sporadic LB epiphytoses as well as for the EB control. The ExelGrow application improves the immune status of plants thus preventing their infection with EB.

**Author contributions:** conceptualization MK; data curation VD; formal analysis MK, NS; investigation AU, AV, VD; methodology MK, VD; project administration MK; supervision MK; validation MK, NS; visualization MK, NS; writing – original draft MK, NS; writing – review and editing MK.

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## Upotreba bioloških i hemijskih fungicida i biostimulatora za zaštitu krompira od alternarije i kasne plamenjače

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Ekološki rizici povezani sa hemijskim pesticidima podstiču primenu biopesticida i integrisano upravljanje štetočinama. Studija je uporedila efikasnost fungicida Manfil i biopreparata Agat-25K (pojedinačnog ili kombinovanog sa EkelGrovi biostimulatorom) protiv kasne (LB) i rane (EB) bolesti krompira. Terenski ogledi su sprovedeni sa tri sorte krompira (Arizona, Vektor, Aluett) različitih nivoa otpornosti na bolesti LB i EB. Razvoj pepelnice registrovan je kao LB (Arizona), LB+EB (Vektor) i EB (Aluett). Agat-25K je odložio razvoj LB u epifitotskim uslovima na osetljivoj cv. Arizona (biološka efikasnost 39%), iako je bila inferiorna u odnosu na Manfil (62%). Naprotiv, za cv. Aluett, Agat-25K je pokazao značajniju supresiju EB od Manfila (96 prema 83%). U poređenju sa kontrolom, povećanje prinosa cv. Arizona je bila 15,8 t/ha za Agat-25K i 25,4 t/ha za Manfil. Tržišnost gomolja porasla je za 28 i 13% za Agat-25K i Manfil, respektivno. Za srednje otporne i otporne sorte (Vektor, Aluett) Agat-25K je dao najveći prinos u poređenju sa Manfilom (19,1 prema 16,7 t/ha za Vektor i 4,5 prema 1,4 t/ha za Aluett, respektivno), dok je tržišnost krtola porasla za 15% (Vektor) i 18% (Aluett). Agat-25K se može preporučiti za upotrebu na umereno rezistentnim sortama pod sporadičnim epifitotijama LB i za kontrolu EB. Prvo se pokazalo da primena EkelGrovi sa Manfilom i Agat-25K poboljšava njihovu biološku efikasnost na sortama umereno otpornim na LB i LB (do 12%).

**Ključne reči:** *Alternaria solani*, biopreparati, integrisana zaštita bilja, krompir, pepelnice na listovima, *Phytophthora infestans*