

<https://doi.org/10.15407/microbiolj85.06.066>

N.Y. PARKHOMENKO, I.K. KURDISH*

Zabolotny Institute of Microbiology and Virology, NAS of Ukraine,
154 Akademika Zabolotnoho Str., Kyiv, 03143, Ukraine

*Author for correspondence; e-mail: ivan.kurdish2016@gmail.com

THE INFLUENCE OF THE COMPLEX BACTERIAL PREPARATION AZOGRAN ON SOME PHYSIOLOGICAL-BIOCHEMICAL PROPERTIES AND PRODUCTIVITY OF POTATO PLANTS INFECTED BY THE POTATO VIRUS X

*The problem of increasing the yield of cultivated plants attracts the attention of specialists and prompts scientists to study viral plant diseases as one of the causes of significant crop losses. A virus is an endogenous parasite that, having entered plant cells, uses the functions of cell organoids and biosystems for its reproduction, which leads to plant disease and a decrease in their productivity. One of the promising approaches to reducing viral damage to plants is the use of microbial antiviral preparations. The complex bacterial preparation Azogran, developed at the Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine, is based on the interaction of phosphate-mobilizing bacteria *Bacillus subtilis* IMV B-7023, nitrogen-fixing bacteria *Azotobacter vinelandii* IMV B-7076, and clay mineral — bentonite. Phosphate-mobilizing bacteria and nitrogen-fixing bacteria significantly improve the growth and development of plants, essentially increasing the yield of agricultural crops. However, the effect of Azogran on the development and yield of potato plants infected with the X-virus has not been sufficiently studied. Taking this into account, the **aim** of the work was to study the effect of the complex bacterial preparation Azogran on the growth, some physiological and biochemical properties, and productivity of potato plants, uninfected and infected by the potato virus X. **Methods.** Microbiological (obtaining the bacterial preparation Azogran, used in granular form (0.25 g)), virological (isolation of potato virus X, inoculation of plants with potato virus X), serological (testing for the presence or absence of the virus), biochemical (determination of chlorophylls a, b and carotenoids), spectrophotometric, and statistical analysis. **Results.** In the conducted experiments, the Azogran was used in granular form in the amount of one and two granules. The effectiveness of the use of the preparation, which stimulates the intensive growth of plants of two varieties of potatoes (early variety Spokusa and medium variety Diva), which leads to an increase in their yield, is shown. The positive effect of Azogran was observed on the development of both infected and non-infected plants. It was found that when two granules of the preparation were applied, the yield of potatoes of the Diva variety increased by 27.02%, and the yield of potatoes of the Spokusa variety under the same conditions increased by 29.46%. A similar effect of Azogran was observed on plants infected with the*

Citation: Parkhomenko N.Y., Kurdish I.K. The Influence of the Complex Bacterial Preparation Azogran on Some Physiologo-Biochemical Properties and Productivity of the Potato Plants Infected by the Potato Virus X. *Microbiological journal*. 2023 (6). P. 66—76. <https://doi.org/10.15407/microbiolj85.06.066>

© Publisher PH «Akademperiodyka» of the NAS of Ukraine, 2023. This is an open access article under the CC BY-NC-ND license (<https://creativecommons.org/licenses/by-nc-nd/4.0/>)

potato virus X, only to a lesser extent: in the Diva variety, the yield was 16.3% higher compared to the control (control — plants not infected by the virus), and in the Spokusa variety it was higher by 22.59%. When studying the content of chlorophylls a and b and carotenoids in virus-infected potato plants, it was established that the content of chlorophylls at different stages of virus infection was different, but the content of chlorophylls and carotenoids was higher in plants bacterized with Azogran. **Conclusions.** The complex bacterial preparation Azogran has a positive effect on the development of uninfected and infected plants, reducing the harmfulness of viral infections and contributing to the increase in potato productivity. Azogran can be successfully used in crop production to increase the quality and quantity of crop yields.

Keywords: complex granular bacterial preparation Azogran, potato virus X, potato plants.

One of the most common agricultural crops is the potato, which is grown in many countries of the world. However, these plants are strongly affected by viral diseases that cause great damage to agricultural production. It is known that in the case of viral damage to potatoes, the loss of its harvest can be 30—40%, and in some cases up to 80—94%. In connection with the deterioration of the ecological situation, which leads to climate change, there is a significant spread of pathogens of potato viral diseases [1, 2]. Therefore, various substances and preparations are used to fight viral infections. Currently, there are many chemicals that inhibit phytovirus infection. But the use of such antiviral preparations contributes only to the reduction of the spread of the infection, and not to its elimination [1]. Moreover, many questions remain to explain the mechanism of antiviral action of chemical compounds, which often have a negative effect not only on viral infection, but also on the growth and development of plants. There are no effective methods of combating viral diseases. The main means of their control in agriculture are the use of preventive measures based on the introduction of varieties with increased natural resistance to pathogens, seed quality control, control of infection vectors, and sanitation of affected fields with the removal of diseased plants [2, 3]. The use of biological preparations can be one of the effective, ecologically safe methods of reducing the harmfulness of viral infections in agro-ecosystems. During the pre-sowing treatment of seeds with microbial preparations of Ukrainian production («Ekovital», «Hetomik», «Rhizofobit», «Rizogumin», and «Haupsin»), a significant decrease in the concentration of the

virus in plants and a decrease in the negative impact of viral infection are observed [3]. In this regard, there is a need to invent biologically active means of suppressing viral infection and stimulating the growth and productivity of plants.

Recent studies have shown that in the fight against viral infections, a promising direction in the field of potato technology is the use of nanotechnology, as well as the combined use of nanoparticles in combination with microbial preparations [4]. Thus, the multi-component microelement preparation «Avatar-2 protection» and the combination of the preparation «Azogran» with the composition of Se+1 nanoparticles significantly reduced the frequency of detection of viral diseases [5].

Considering this fact, a promising approach is the study of the effect of the complex bacterial preparation Azogran on the growth and productivity of potato plants infected by the potato virus X (PVX). This preparation was proposed by the team of the authors of the Institute of Microbiology and Virology of the National Academy of Sciences of Ukraine [6]. The preparation Azogran is created on the basis of the interaction of phosphate-mobilizing bacteria *Bacillus subtilis* IMV B-7023 [7], nitrogen-fixing bacteria *Azotobacter vinelandii* IMV B-7076 [8], with clay mineral bentonite [9]. It was previously established that the strain *B. subtilis* IMV B-7023, which is part of the Azogran preparation, is characterized by a wide spectrum of antagonistic activity against phytopathogenic bacteria and micromycetes [10]. *A. vinelandii* IMV B-7076 also has the ability to suppress the spread of phytopathogenic micromycetes [11]. Previous studies have estab-

lished the effectiveness of the antiviral effect of one-, two-, and three-day culture fluid of *Bacillus subtilis* IMV B-7023 on the infectiousness of tobacco mosaic virus (TMV) *in vitro*, as well as the antiviral activity of prophylactic (preventive) treatment of dope seeds [12], but the effect of the preparation Azogran on the course of viral infection in plants has not been studied enough.

The **aim** of this work was to study the effect of the complex granular bacterial preparation Azogran on the growth, development, and yield of potatoes, and some changes in the biochemical indications of plants after their infection by the potato virus X.

Materials and methods. In the conducted investigations, the granular bacterial preparation Azogran was used. The action of the preparation was studied on uninfected and infected by PVX potato plants of the following varieties: Spokusa (early variety) and Diva (medium variety). Potatoes of the Diva variety (Seedera) were sown from seeds and used 3 years after sowing. Potatoes of the Spokusa variety were bought in the Zhytomyr region. Small-area experiments were conducted on loamy soil of the Zhytomyr region. Predecessors were vegetable crops: peppers, garlic, and onions. After harvesting, mustard was sown in autumn. For each version of the experiment, 10 potato plants were planted in triplicate. The experiments were carried out during three years (2020—2022). The following variants of each potato variety were used in the experiments: a) uninfected potato plants without Azogran (control); b) introduction of 1 granule of Azogran (0.25 g) along with potatoes; c) 2 granules of Azogran (0.5 g); d) plants infected by PVX without applying Azogran; e) plants infected by PVX + 1 granule of Azogran; f) plants infected by PVX + 2 granules of Azogran.

Potatoes were planted in the first decade of May. One and a half months after planting potatoes, in the stage of budding, the plants were mechanically infected by the potato virus X. For this purpose, the potato leaves that were previously dusted with

abrasive powder (to improve the penetration of the virus into plant tissues) were applied with the help of a brush to the viral material at a concentration of 1 mg/mL. X-virus, kindly provided by the Department of Plant Viruses, was isolated from infected *Datura stramonium* plants [13]. The plants of *Datura stramonium* are widely used as a host plant for PVX, i.e., it is an indicator plant for PVX. *Datura stramonium* is immune to potato Y-, S-, and M-viruses and hypersensitive to TMV, responds to infection by the X-virus with clear mosaic symptoms, which appear, as a rule, on the 12—14th days. With a decrease in temperature, the symptoms are manifested by necrotic lesions, and at an elevated temperature, the presence of a mosaic can be masked [14]. With the help of anti-serum to PVX, plants were checked for the presence of the virus in them.

It is known that viral infection leads to a low level of photosynthetic pigments [15], it was therefore interesting to investigate changes in the amount of pigments during plant growth. To determine the content of pigments chlorophylls *a*, *b* and carotenoids, leaves of different variants of potato plants were selected on the 3rd, 7th, 10th, and 14th days after their inoculation by the virus X, as well as in control variants. On the same day after selection, the collected leaves were washed and dried, and a sample was taken from each studied sample to determine the content of pigments in the leaves of infected and uninfected potato plants. The pigments were extracted with 96% ethanol in the presence of magnesium carbonate to neutralize the acids of the cell juice and to prevent the disintegration of the pigments [16]. The optical density of ethanol extracts after filtration was determined using an SF-48 spectrophotometer by the three-wavelength method, determining the optical density (*E*) of the extract at wavelengths of 665, 649, and 441 nm, which corresponds to the absorption maximum of chlorophyll *a*, chlorophyll *b*, and carotenoids, respectively [17]. The concentration of chlorophylls *a* and *b* (*C_a* and *C_b*) was calculated ac-

ording to the equations:

$$Ca = 13.70 \times E_{665} - 5.76 \times E_{649} \text{ (mg/L);}$$

$$Cb = 25.80 \times E_{649} - 7.60 \times E_{665} \text{ (mg/L).}$$

The concentration of carotenoids (Cc) in the total extract of pigments was calculated according to the equation:

$$Cc = 4.7 \times E_{441} - 0.27 \times C \times (a+b) \text{ (mg/L),}$$

where E_{665} , E_{649} , and E_{441} are the optical density of the extract at a wavelength of 665, 649, and 441 nm, respectively; C is the concentration of pigments in the extract, mg/L;

Having determined the concentration of the pigment in the extract, its content in the examined fabric was calculated according to the formula:

$$F = (V \times C) / P,$$

where F is the pigment content in the plant material (mg/g of raw substance);

V is the hood volume (L); C is the pigment concentration; P is the weight of plant material (g). The obtained results were processed statistically [18].

Results. It is known from the literature that viruses influence the physiological and biochemical processes in infected plants. Plants affected by viruses are characterized by severe metabolic

changes and typical symptoms such as chlorosis and leaf blade necrosis. In particular, with chlorosis, the color of chloroplasts changes from green to yellow or white in the form of a mosaic, and with necrotizing, cell death generally occurs, which leads to blackening of the leaf surface. Thus, virus damage can make plant leaves shriveled or curled, with pronounced chlorosis or necrosis. This reduces the ability of plants to photosynthesize, ultimately leading to a low level of photosynthetic pigments (chlorophylls and carotenoids) [15, 19].

The research results on the determination of the content of chlorophylls *a*, *b* and carotenoids in uninfected potato plants showed that after Azogran was added to uninfected potato plants, the quantitative content of chlorophylls and carotenoids was significantly higher than in uninfected potato plants without Azogran (Table 1). Thus, when two granules of Azogran were placed in a hole when planting potatoes of the Diva variety, the content of chlorophyll *a* was 1.82 mg/g, chlorophyll *b* — 1.25 mg/g, and carotenoids — 1.24 mg/g, compared to plants without Azogran, in which the values of these indicators were 1.12 mg/g and 0.51 mg/g for chlorophylls *a*, *b* and 0.67 mg/g for carotenoids.

When Azogran granules were added to potato plants of the Spokusa variety, the results were even better: the content of chlorophyll *a* was 1.81 mg/g, *b* — 2.39 mg/g, and carotenoids —

Table 1. Determination of chlorophylls *a* and *b* and carotenoids in non-infected potato plants using Azogran

Research options	Quantitative content of pigments, mg/g		
	Chlorophyll <i>a</i>	Chlorophyll <i>b</i>	Carotenoids
Potatoes of the Diva variety			
Potato plants without Azogran	1.12 ± 0.14	0.51 ± 0.09	0.67 ± 0.07
Potato plants +1 Azogran granule	1.71 ± 0.04	1.2 ± 0.06	0.84 ± 0.05
Potato plants +2 granules of Azogran	1.82 ± 0.06	1.25 ± 0.06	1.24 ± 0.07
Potatoes of the Spokusa variety			
Potato plants without Azogran	1.25 ± 0.10	0.7 ± 0.06	0.45 ± 0.06
Potato plants +1 Azogran granule	1.45 ± 0.13	1.55 ± 0.07	0.73 ± 0.10
Potato plants +2 granules of Azograna	1.81 ± 0.09	2.39 ± 0.10	0.95 ± 0.11

0.95 mg/g, compared to uninfected potato plants without Azogran, in which the content of chlorophyll *a* was 1.25 mg/g, chlorophyll *b* — 0.7 mg/g, and carotenoids — 0.95 mg/g (Table 2).

In relation to infected potato plants with the addition of Azogran, the content of chlorophylls *a*, *b* and carotenoids has been higher compared to infected and uninfected plants without the addition of Azogran. For example, on the 7th day after infection in infected plants + 2, granules of Azogran, the content of chlorophylls *a*, *b* was 1.62 mg/g and 0.69 mg/g, while without Azogran these indicators reached values of 0.92 mg/g and 0.45 mg/g. A similar pattern was observed for the content of carotenoids — 1.01 mg/mL in in-

fecting plants with Azogran and 0.55 mg/mL in infected plants without the addition of Azogran.

The studies of the content of chlorophylls *a*, *b* and carotenoids in infected potato plants of the Spokusa variety at different stages of the infection showed that the highest value of chlorophylls *a*, *b* was observed on the 3rd day after infection (*a* — 1.5 mg/g, *b* — 1.09 mg/g); on the 7th day (*a* — 1.58 mg/g, *b* — 1.78 mg/g) in comparison with control plants on the 3rd day (*a* — 1.25 mg/g, *b* — 0.7 mg/g); on the 7th day (*a* — 1.25 mg/g, *b* — 1.4 mg/g) (Table 3).

The content of carotenoids in virus-infected plants on the 3rd (1.21 mg/g) and the 7th day (1.33 mg/g) was also higher compared to the

Table 2. Determination of chlorophylls *a*, *b* and carotenoids in potato plants of the Diva variety infected by PVX at different stages of the infection development

Research options	Quantitative content of pigments, mg/g		
	Chlorophyll <i>a</i>	Chlorophyll <i>b</i>	Carotenoids
The 3 rd day after infection by PVX			
Uninfected plants (control)	1.12 ± 0.14	0.51 ± 0.09	0.67 ± 0.07
Infected potato plants	1.12 ± 0.12	0.58 ± 0.06	0.78 ± 0.07
Infected plants + 1 granule of Azogran	1.26 ± 0.05	0.66 ± 0.10	0.8 ± 0.06
Infected plants + 2 granules of Azogran	1.39 ± 0.03	0.75 ± 0.08	0.88 ± 0.05
The 7 th day after infection by PVX			
Uninfected plants (control)	1.25 ± 0.10	0.61 ± 0.08	0.75 ± 0.06
Infected potato plants	0.92 ± 0.08	0.45 ± 0.10	0.55 ± 0.07
Infected plants + 1 granule of Azogran	1.25 ± 0.06	0.72 ± 0.08	0.87 ± 0.05
Infected plants + 2 granules of Azogran	1.62 ± 0.07	0.69 ± 0.07	1.01 ± 0.12
The 10 th day after infection by PVX			
Uninfected plants (control)	0.06 ± 0.01	0.93 ± 0.08	0.81 ± 0.07
Infected potato plants	0.08 ± 0.01	0.08 ± 0.10	0.74 ± 0.06
Infected plants + 1 granule of Azogran	0.21 ± 0.60	0.61 ± 0.07	0.45 ± 0.09
Infected plants + 2 granules of Azogran	0.32 ± 0.07	0.84 ± 0.06	0.84 ± 0.1
The 14 th day after infection by PVX			
Uninfected plants (control)	0.14 ± 0.09	0.72 ± 0.07	0.49 ± 0.06
Infected potato plants	0.25 ± 0.07	0.46 ± 0.10	0.57 ± 0.06
Infected plants + 1 granule of Azogran	0.27 ± 0.06	0.79 ± 0.11	0.68 ± 0.05
Infected plants + 2 granules of Azogran	0.31 ± 0.08	0.91 ± 0.07	0.82 ± 0.12

control plants on the 3rd (0.45 mg/g) and the 7th (0.79 mg/g) day. It should be noted that when infected by the virus and added with Azogran, the pigment content was higher in all cases than in uninfected and infected plants without Azogran.

In infected potato plants of the Spokusa variety, an increase in the content of chlorophylls and carotenoids was observed from the 3rd day of infection in comparison with control plants and plants treated with Azogran (Table 3). The content of carotenoids in virus-infected plants on the 3rd day (1.21 mg/g) and on the 7th day (1.33 mg/g) was also higher compared to the control plants (on the 3rd day — 0.45 mg/g and on the 7th day — 0.79 mg/g).

It should be noted that an increase in the content of chlorophylls and carotenoids of infected plants was observed from the 3rd day of infection in potato plants of the Spokusa variety compared to uninfected plants and plants to which Azogran was added. An increase in the pigment content in infected plants was observed up to the 10th day. On the 10th day, the pigment content in those infected with the virus decreased, and on the 14th day after infection, it increased slightly. Although it can be seen that the presence of Azogran in infected plants significantly improves their condition compared to infected plants and potato plants without the use of Azogran.

Table 3. Determination of chlorophylls *a*, *b* and carotenoids in PVX-infected potato plants of the Spokusa variety at different stages of infection development

Research options	Quantitative content of pigments, mg/g		
	Chlorophyll <i>a</i>	Chlorophyll <i>b</i>	Carotenoids
The 3 rd day after infection by PVX			
Uninfected plants (control)	1.25 ± 0.10	0.7 ± 0.06	0.45 ± 0.06
Infected potato plants	1.50 ± 0.11	1.09 ± 0.06	1.21 ± 0.07
Infected plants + 1 granule of Azogran	1.40 ± 0.07	1.32 ± 0.07	1.04 ± 0.12
Infected plants + 2 granules of Azogran	1.30 ± 0.10	0.92 ± 0.09	0.99 ± 0.07
The 7 th day after infection by PVX			
Uninfected plants (control)	1.25 ± 0.06	1.40 ± 0.08	0.79 ± 0.06
Infected potato plants	1.58 ± 0.08	1.78 ± 0.06	1.33 ± 0.07
Infected plants + 1 granule of Azogran	1.06 ± 0.08	1.16 ± 0.08	0.68 ± 0.06
Infected plants + 2 granules of Azogran	1.30 ± 0.10	1.43 ± 0.07	0.81 ± 0.09
The 10 th day after infection by PVX			
Uninfected plants (control)	0.13 ± 0.07	0.78 ± 0.06	0.69 ± 0.09
Infected potato plants	0.09 ± 0.10	0.78 ± 0.11	0.67 ± 0.06
Infected plants + 1 granule of Azogran	0.13 ± 0.07	0.77 ± 0.05	0.71 ± 0.08
Infected plants + 2 granules of Azogran	0.36 ± 0.06	0.82 ± 0.09	0.81 ± 0.10
The 14 th day after infection by PVX			
Uninfected plants (control)	0.13 ± 0.09	0.78 ± 0.07	0.68 ± 0.06
Infected potato plants	0.17 ± 0.06	0.87 ± 0.08	0.70 ± 0.08
Infected plants + 1 granule of Azogran	0.15 ± 0.06	0.69 ± 0.10	0.56 ± 0.09
Infected plants + 2 granules of Azogran	0.25 ± 0.07	0.82 ± 0.12	0.81 ± 0.11

The bacterial preparation Azogran also influences the development of green shoots of potatoes. The research results show that infected plants of both potato varieties had a less developed and less branched stem system compared to uninfected plants.

However, when Azogran was added to potato plants infected by the virus, the morphological indicators of the plants were significantly better than those of plants without the introduction of the bacterial preparation (Table 4).

The development of a powerful and developed stem system with the use of the preparation Azogran significantly increased the yield of potatoes (Table 5). When one granule of Azogran was applied, the yield of potatoes increased by 13.29%, and when two granules were applied, it increased by 27.02% for the Diva variety and from 16.00 to 29.46% for the Spokusa variety, compared to

potato plants where Azogran was not applied. The effect of Azogran was more pronounced on Spokusa variety potato plants.

Thus, the obtained results indicate that viral infection of potato plants causes a decrease in the amount of the crop. The use of the bacterial preparation Azogran improves the development of plants affected by viruses, protecting them from the effects of viral infection and increasing the yield of potatoes.

Discussion. It is known from the literature that viruses affect the majority of physiological and biochemical processes in infected plants, including the activity of chloroplasts [15, 19, 20]. In particular, chloroplasts and photosynthesis are widely recognized as common targets for many plant viruses. As a result, viral infection leads to low levels of photosynthetic pigments (chlorophylls and carotenoids) [15]. The viral in-

Table 4. The effect of Azogran on the stem length of potato plants of the Diva and Spokusa varieties

Research options	Average stem length of potato plants (cm)	Percent to control%	Average stem length of potato plants (cm)	Percent to control%
	Potato variety Diva		Potato variety Spokusa	
Uninfected plants (control)	64.55 ± 2.89	100	69.02 ± 1.01	100
Infected potato plants	57.40 ± 6.08	86.25	67.46 ± 2.32	97.74
Infected plants + 1 granule of Azogran	62.45 ± 3.61	93.84	76.77 ± 1.55	110.11
Infected plants + 2 granules of Azogran	65.76 ± 2.35	101.87	77.94 ± 1.07	112.07

Table 5. The effect of Azogran on the yield of potato plants of the Diva and Spokusa varieties*

Research options	Total amount of potato harvest (kg/10 bushes)	Percentage to control, %	Total amount of potato harvest (kg/10 bushes)	Percentage to control, %
	Potato variety Diva		Potato variety Spokusa	
Control plants	9.18 ± 1.12	100.00	9.3 ± 0.58	100.00
Control plants +1 Azogran granule	10.4 ± 1.27	113.29	10.82 ± 0.33	116.00
Control plants +2 Azogran granule	11.66 ± 1.39	127.02	12.04 ± 0.57	129.46
Plants infected by PVX	7.66 ± 1.03	83	7.08 ± 0.47	76.12
Plants infected by PVX + 1 granule of Azogran	8.22 ± 1.01	89.54	7.92 ± 0.48	85.16
Plants infected by PVX + 2 granules of Azogran	8.72 ± 0.06	95.09	9.18 ± 0.50	98.71

*The data presented in the Table were obtained during 3 years.

fection changes the state of chloroplasts, turning their color from green to yellow or white in the form of a mosaic [19, 20]. During the period of maximum replication of turnips' yellow mosaic virus, changes are observed in the leaves, which apparently reflect the specific effect of its replication on chloroplasts [19].

The process of photosynthesis is one of the most important links of plant metabolism. It is very sensitive to growing conditions. It was established that the most intensive synthesis of green pigments, namely chlorophyll *b*, occurs in the phase of leaf closure in interrows: when leaves in rows are closed, chlorophyll *a* is synthesized more intensively. The least intensive synthesis of chlorophyll *a* is at the initial stages of plant growth and development, due to the small area of the leaf surface and the low level of PHAR assimilation [21]. Thus, in the first half of the vegetation of beet plants, when 4 pairs of leaves have formed, the synthesis of chlorophyll *b* prevails, and in the second phase of technical maturity, the synthesis of chlorophyll *a* prevails [21].

As shown in Table 2, the indicators of the number of chlorophylls and carotenoids in potato plants of the Diva variety on the 3rd day after infection are almost the same compared to uninfected plants, and only on the 7th day after infection, the number of chlorophylls *a*, *b* and carotenoids significantly decreased and amounted to 0.92 mg/g and 0.45 mg/g for chlorophylls *a*, *b* and to 0.55 mg/g for carotenoids compared to uninfected plants. In uninfected plants, the content of chlorophyll *a* was 1.25 mg/g, *b* — 0.61 mg/g, and carotenoids — 0.75 mg/g. On the 10th day, the amount of chlorophyll *a* in infected plants begins to increase, and on the 14th day, it almost doubles. Carotenoids also increase compared to the control.

As for the Spokusa variety of potato plants, there is observed an increase in chlorophylls *a*, *b* and carotenoids already on the 3th day after infection, reaching a maximum on the 7th day: chlorophyll *a* — 1.58 mg/g, *b* — 1.78 mg/g, and

carotenoids — 1.33 mg/g compared to the control, where the content of chlorophyll *a* reached 0.1 mg/g, chlorophyll *b* — 0.78 mg/g, and carotenoids — 0.69 mg/g. A decrease in the content of chlorophylls *a*, *b* and carotenoids was observed on the 10th day, and on the 14th day, these indicators increased compared to the control (Table 3).

According to the literature data, the main function of chloroplasts is the process of photosynthesis to obtain energy for the plant (in the form of ATP). In addition to this function, chloroplasts perform many others, including the synthesis of amino acids, nucleotides, and many others. It is known that the reproduction of potato virus X ends, mainly, by the 14th day [14]. Perhaps, during this period, the most energy is used for the maturation and release of mature viral particles. Because of this, an increase in the content of chlorophylls and carotenoids in infected potatoes of the Diva variety is observed to be 0.08 mg/g for chlorophyll *a* on the 10th day, 0.25 mg/g for chlorophyll *a*, and 0.57 mg/g for carotenoids on the 14th day compared to the control: 0.06 mg/g, 0.14 mg/g, and 0.49 mg/g, respectively (Table 1).

Regarding the infected potato plants with added Azogran, the indicators of the content of chlorophylls and carotenoids were higher compared to the control and infected plants without the use of Azogran. As can be seen from the obtained data, the amount of chlorophyll *a*, *b* and carotenoids changes under the influence of viral infection, moreover, the changes are observed depending on the stage of virus reproduction in the cell. Obviously, in different varieties of potatoes, the interaction between the virus and the plant-host is expressed differently. Plant parameters of Spokusa potato plants, such as shoot length and yield, were significantly higher compared to Diva potato plants. The same results were observed in uninfected potato plants, both with and without the application of Azogran. In our opinion, this already depends on the variety: potato bushes of the Diva variety are more compact and low, whereas potato bushes of the

Spokusa variety are more branched and tall. Therefore, the results were compared only with the control of one variety, since different varieties of plants have their own differences when interacting with the virus. It is possible that the protective reactions of the host plant and the virulence of the virus play a significant role in the development of a viral infection.

Thus, improving the conditions for the growth and development of plants when they are infected by viruses, makes it possible for plants to resist viral infection and promotes more active adaptation to the action of adverse external factors. The results of these studies indicate a significant effect of the complex bacterial preparation Azogran on the growth, development, and productivity of both healthy potato plants and their viral infection.

The results of our research are consistent with the data obtained by other authors, who proved the effectiveness of the use of the microbial preparation Azogran for combating bacterial, fungal, and viral diseases of potatoes, increasing the marketability of tubers and yield. The authors believe that the composition of Se+1 nanoparticles and the preparation Azogran positively affected the microbiota of the potato rhizosphere and the physiological state of the plants [4, 5]. When Azogran is introduced into the root system of vegetable and technical crops, the nitrogen and phosphorus nutrition of plants improves, their growth and development are significantly stimulated, and productivity increases by 18–37% [22].

The obtained results indicate that the use of the complex biological preparation Azogran improves the growth and development of plants that are affected by viruses. For example, in uninfected potato plants of the Diva variety, the content of chlorophyll *a* was 1.12 mg/g, chlorophyll *b* — 0.51 mg/g, and carotenoids — 0.67 mg/g. After adding two Azogran granules to this agroecosystem, the content of chlorophyll *a* was 1.82 mg/g, chlorophyll *b* — 1.25 mg/g, and carotenoids —

1.24 mg/g. When potato plants were infected by the virus X and Azogran was added, the contents of chlorophylls *a*, *b* and carotenoids were significantly higher at different stages of the infection. This fact was also noted during the study of the content of these pigments in potato plants of the Spokusa variety. In the control potato plants, the content of chlorophyll *a* was 1.25 mg/g, chlorophyll *b* — 0.7 mg/g, carotenoids — 0.45 mg/g, while the content of chlorophyll *a* increased up to 1.81 mg/g when two granules of Azogran were added to the control plants, chlorophyll *b* — 2.39 mg/g, carotenoids — 0.95 mg/g. When potato plants were infected by the virus, the content of pigments was significantly higher when using the preparation Azogran, as evidenced by the data obtained.

Conclusions. The application of the complex biological preparation Azogran improves the growth and development of potato plants under normal growth conditions, as well as under viral infection.

The use of the bacterial preparation Azogran increased by 27.02% the yield of potatoes in the Diva variety, and by 29.46% in the Spokusa potato variety. With the introduction of the preparation Azogran (0.5 g) in virus-infected plants of the Diva variety, the yield increased by 16.3%, and in the Spokusa variety — by 22.59%, compared to infected plants without Azogran.

The obtained results show that the use of the complex bacterial preparation Azogran has a positive effect on the development of plants, increasing the yield of both uninfected and PVX-infected potato plants. The increase in the area of the above-ground mass affects the assimilation of nutrients, contributes to the intensification of the processes of photosynthesis in plants, and increases their yield.

Acknowledgements. The authors are very grateful to the head of the Department of Plant Viruses A.M. Kyrychenko and to all employees of this department for consulting, providing viral material, and facilitating our work.

REFERENCES

1. Volkova IV, Reshotko LM, Dmytruk OO. Spread of viral disease pathogens in culture cultivation zones. *Silskohospodarska mikrobiologhiia. Agricultural Microbiology*. 2020; 32:67—73.
2. Desk encyclopedia of plant and fungal virology. M.H.van Regenmortel, B.W. Mahy, editors. Oxford. Academic Press. 2009. p. 633.
3. Kyrychenko AM. Biological methods of combating plant viruses. Special edition of the offer. Bioprotection and biological preparations are an actual perspective. 2017. pp. 42—46.
4. Vasylenko A, Derevianko S. The use of nanoparticles and nanotechnologies in potato growing. *Herald of Agrarian Science. Crop production, fodder production*. 2022; 9(834):43—54.
5. Vasylenko AV. The effect of nanoparticles of metals and non-metals, the multicomponent microelement preparation «Avatar-2 protection» and the microbial preparation «Azogran» on the degree of damage to potatoes by infectious diseases and the frequency of their detection. *Agroecological Journal*. 2021; 4:90—97.
6. Kurdish IK, Roy AO, Titova LV. [Granulated preparations of complex action based on nitrogen-fixing and phosphate-mobilizing bacteria]. *Agrarian education on the beginning of the third millennium. Materials of International scientific-practical conference, September 18—21, Lviv*. 2001; 189—194. Ukrainian.
7. Patent of Ukraine № 54923A [The bacterial strain *Bacillus subtilis* for bacterial fertilizer for crop production]. Kurdish IK, Roy AA. Publ. 2003. Bul. №3. Ukrainian.
8. Patent of Ukraine № 72856 [Strain bacteria *Azotobacter vinelandii* for bacterial fertilizer obtaining for plant-growing]. Kurdish IK, Bega ZT. Publ. 2006. Bul. № 8. Ukrainian.
9. Declarative patent of Ukraine No.57269. [The method of obtaining granular bacterial preparations]. Kurdish IK, Roy AA, Bega ZT. Publ. 2003. Bul. № 6. Ukrainian.
10. Roy AA, Pasichnik LA, Tserkovnyak LS, Khodos SF, Kurdish IK. [Influence of bacteria of the genus *Bacillus* on causative agents of tomato bacterial cancer]. *Mikrobiol Z*. 2012; 74(5):74—80. Ukrainian.
11. Chuiko NV, Chobotarov AYu, Savchuk YaI, Kurchenko IM, Kurdish IK. Antagonistic activity of *Azotobacter vinelandii* IMV B-7076 against phytopathogenic microorganisms. *Mikrobiol Z*. 2020; 82(5):21—29.
12. Skorochoch IO, Roy AO, Kyrychenko AM. Antiviral affect of cultural liquid of *Bacillus subtilis* IMV B-7023 on *Datura stramonium* lives infected with tobacco mosaic virus. *Biotechnology for agriculture and environmental protection*. 2016; 213—214. Odesa, Ukraine.
13. Maksymenko LO, Parkhomenko NY. *In vitro* and *in vivo* phosphorylation of a coat protein of potato virus X. *Mikrobiol Z*. 2021; 83(5):76—81.
14. Grama DP, Parkhomenko NY, Krayev VG. Development of systematic infection of potato virus X in the *Datura stramonium* L. plants. *Mikrobiol Z*. 1981; 43(6):780—785.
15. Li X, Cui H, Cui X, Wang A. Altered photosynthetic mechanism during infection with compatible viruses. *Curr Opin Virol*. 2016; 17:19—24.
16. Lichtenthaler HK. Chlorophylls and carotenoids: Pigments of photosynthetic biomembranes. *Methods in Enzymology*. 1987; 148:350—382.
17. Grynenko UV, Zhuravel IO. Determination of the chlorophylls and carotenoids content in spinach leaves (*Spinacia oleracea* L.) Proceedings of scientific work of NMAPO im P.L.Shupyk. 2017; 28:29—33.
18. Lakin GF. [Biometry]. Moscow: Vysshiaia shkola. 1990; 352. Russian.
19. Melnychuk MD. *Fitovirusologhiia: navchalnyyposibnyk [Phytovirusology: trainingmanual]*. Kyiv: Poligraph Konsal'tynh. 2005. Ukrainian.
20. Matthews REF. *Fundamentals of plant virology*. New York: Acad Press. 1992; 403.
21. Prysiashniuk OI, Korovko II. Dynamics of chlorophyll content in sugar beet leaves. *Crop production*. 2013; Latest agricultural technologies.
22. Kurdish IK. Introduction of microorganisms in the agroecosystem. «Naukova Dumka» publishing house of the D.K. Zabolotny Institute of Microbiology and Virology of the NASU. Kyiv. 2010; 180—190.

Received 23.04.2023

Н.Й. Пархоменко, І.К. Курдиш

Інститут мікробіології і вірусології ім.Д.К. Заболотного НАН України,
вул. Академіка Заболотного, 154, Київ, 03143, Україна

ВПЛИВ КОМПЛЕКСНОГО БАКТЕРІАЛЬНОГО ПРЕПАРАТУ АЗОГРАН НА ДЕЯКІ ФІЗІОЛОГО-БІОХІМІЧНІ ВЛАСТИВОСТІ І ПРОДУКТИВНІСТЬ КАРТОПЛІ, ІНФІКОВАНОЇ Х-ВІРУСОМ КАРТОПЛІ

Проблема підвищення врожайності культурних рослин привертає увагу спеціалістів і спонукає вчених до вивчення вірусних хвороб рослин як однієї з причин значних втрат урожаю. Вірус — це ендегенний паразит, який потрапивши до клітин рослини, використовує функції клітинних органелів і біосистем для свого відтворення, що призводить до захворюваності рослин і зниження їх продуктивності. Одним із перспективних підходів для зниження вірусного ураження рослин є застосування мікробних препаратів антивірусної дії. Комплексний бактеріальний препарат Азогран, розроблений в Інституті мікробіології і вірусології НАН України, базується на взаємодії фосфатмобілізуючих бактерій *Bacillus subtilis* IMV B-7023, азотфіксувальних бактерій *Azotobacter vinelandii* IMV B-7076 і глинистого мінерала — бентоніта. Фосфатмобілізуючі бактерії і азотфіксувальні бактерії значно покращують ріст і розвиток рослин, суттєво підвищують урожай сільськогосподарських культур. Однак вплив Азограну на розвиток і урожайність рослин картоплі, інфікованих Х-вірусом, недостатньо вивчено. Зважаючи на це, метою роботи було дослідити вплив комплексного бактеріального препарату Азогран на ріст, деякі фізіолого-біохімічні властивості та продуктивність рослин картоплі, неінфікованих та інфікованих Х-вірусом картоплі. **Методи.** Мікробіологічні (отримання бактеріального препарату Азогран, використаного в гранульованій формі (0,25 г)), вірусологічні (виділення Х-вірусу картоплі, інокуляція рослин Х-вірусом картоплі, серологічне тестування на присутність чи відсутність вірусу), біохімічні (визначення хлорофілів *a*, *b* і каротиноїдів), спектрофотометричні, і статистичний аналіз. **Результати.** У проведених експериментах був використаний препарат Азогран у гранульованому вигляді в кількості однієї та двох гранул. Показана ефективність застосування препарату, який стимулює інтенсивний ріст рослин двох сортів картоплі (раннього сорту Спокуса та середнього сорту Діва), що призводить до підвищення їх урожайності. Позитивну дію Азограну спостерігали на розвитку як інфікованих, так і неінфікованих рослин. Встановлено, що при внесенні двох гранул препарату урожайність картоплі сорту Діва зростала на 27,02 %, а урожайність картоплі сорту Спокуса за тих же умов — на 29,46 %. Аналогічну дію препарату спостерігали і на рослинах, інфікованих Х-вірусом картоплі, тільки в меншій мірі: у сорту Діва урожайність була на 16,3% вищою в порівнянні з контролем (контроль — неінфіковані вірусом рослини), а в сорту Спокуса — вища на 22,59 %. При дослідженні вмісту хлорофілів *a* і *b* та каротиноїдів в інфікованих вірусом рослинах картоплі було встановлено, що вміст хлорофілів на різних стадіях вірусної інфекції був різним, але вміст хлорофілів та каротиноїдів був вищим в рослинах, бактеризованих препаратом Азогран. **Висновки.** Комплексний бактеріальний препарат Азогран позитивно впливає на розвиток неінфікованих та інфікованих рослин, знижуючи шкодочинність вірусних інфекцій та сприяючи підвищенню продуктивності картоплі. Препарат Азогран може з успіхом застосовуватися в рослинництві для підвищення якості та кількості врожаю сільськогосподарських культур.

Ключові слова: комплексний гранульований бактеріальний препарат Азогран, Х-вірус картоплі, рослини картоплі.