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THE INDUCTION OF *TRITICUM AESTIVUM* L. TOLERANCE TO *SEPTORIA TRITICI* BY OXALIC ACID

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Abstract. The influence of fungus *Septoria tritici* on winter wheat (*Triticum aestivum*) cultivars *'Polis´ka 90'* and *'Stolychna'* yield formation and plant development, as well as protective role of oxalic acid were studied in the field experiment. It was shown that treatment by oxalic acid decreases the infected square of flag leaves, ears and stems in both varieties that normalized plant development and reduced the yield loss. The usage of elicitors is the way to induce the plant immunity of cereals and minimize the pesticide pollution of the environment.

Key words: *Septoria tritici, Triticum aestivum*, Septoria tritici blotch, oxalic acid, morphometry, grain, stem, last leaves

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Introduction

Pathogenic fungus *Septoria tritici* Berk. et M.A. Curtis is one of the major causes of losses (10 to 50%) of wheat yield in Ukraine. As an alternative of fungicide usage there are biological methods of plant protection which induce systemic resistance. This type of resistance is based on induction of many genes expression by substances pathogens – biotic elicitors, and therefore it is non-specific (Шакирова 2001). This allows preservation of resources, compared to the cost of the development of new genetically resistant cultivars. Septoria tritici blotch is caused by the fungus *S. tritici*. It is distributed in all wheat-growing areas of the world and is a serious problem in many regions (Захаренко *и др.* 2003). Septoria tritici blotch is the most damaging when attacks the upper leaves and heads of susceptible varieties late in the season.

Effective protection of wheat plants against Septoria tritici blotch is an actual problem of modern biology. It is important to identify Septoria tritici blotch before spraying with a fungicide because nutritional disorders such as aluminum toxicity can be confused with Septoria tritici blotch. Nowadays biogenic elicitors are one of the most perspective approaches decreasing crop losses under fungal infection, and they induce the systemic plant immunity against fungal pathogens (Дмитриев *и др.* 2005; Жук *та ін.* 2013; Жук і Дмитрієв 2013). The studies of oxalic acid role in plant disease resistance are a hot spot of modern cell biology (Toal & Jones 1999). Oxalic acid is known as the general physiological trait. The most of brown-rot basidiomycetes, including *Fomitopsis palustris* (Berk. et M.A. Curtis) Gilb. et Ryvarden accumulate oxalic acid at greater concentrations in culture fluid, whereas white-rot ones do not accumulate because they metabolize and/or decompose oxalic acid by various mechanisms. Nevertheless, the white-rots were observed to accumulate Ca-oxalate during wood decay processes (Kayashima & Katayama 2002).

The plant synthesizes of oxalic acid must coordinate the rate of synthesis with Ca, its accumulation and crystal development (Franceschi & Nakata 2005). There are number of potential pathways for oxalic acid biosynthesis in plant. This organic acid can be formed through the oxidation of glycolate and glyoxylate by the activity of glycolate oxidase (Dumas *et al.* 1995; Cessna *et al.* 2000). These potential substrates can be formed as a byproduct of photorespiration in photosynthetically active tissues, and glycolate oxidase, a peroxisomal enzyme, is

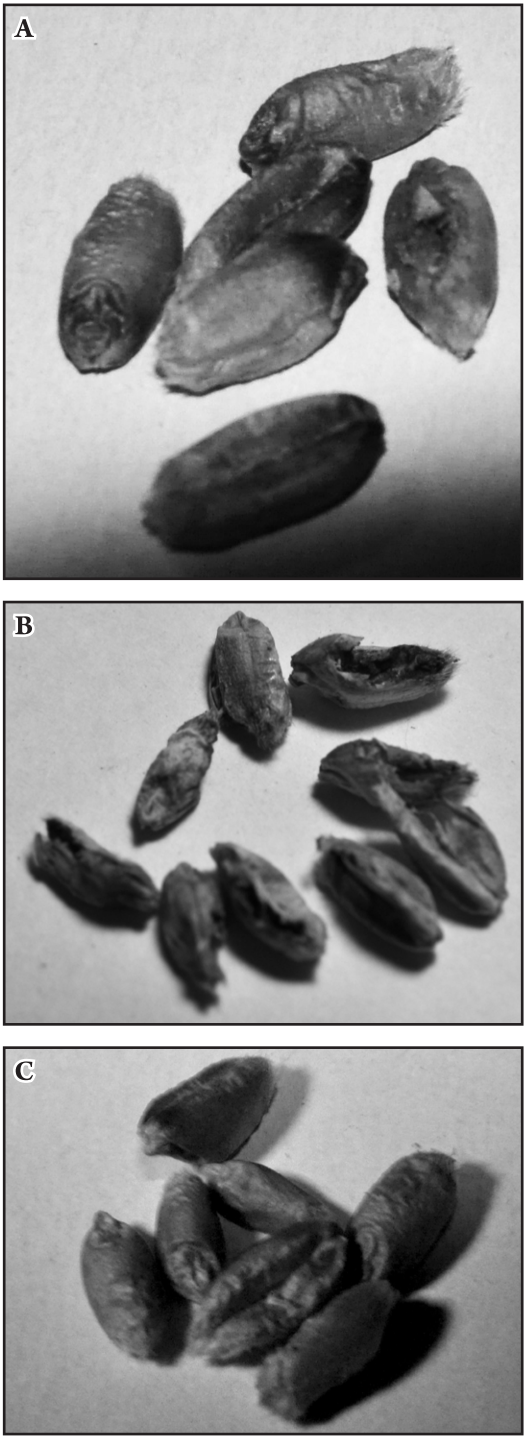

Fig. 1. The grains of winter wheat cultivar *'Polis´ka 90'*: **A** – control; **B** – infected by winter wheat leaf blotch; **C** – infected by winter wheat leaf blotch under oxalic acid treatment.

fairly abundant in green tissues. Oxalate can also be produced by the activity of isocitrate lyase on isocitrate, and through oxidation of oxaloacetate, although the enzyme is not known and this has only been shown in a few tissues. Finally, l-ascorbic acid is a substrate for oxalate synthesis in a number of plant species, but the enzyme(s) responsible for this pathway has not been identified.

The aim of our research was to investigate the role of oxalic acid in wheat plant tolerance to Septoria tritici blotch in field experiment.

Material and methods

The winter wheat (*Triticum aestivum* L.) cultivars *'Polis´ka 90'* and *'Stolychna'* were grown in field conditions in Kyiv district with the standard agriculture technique. The soil was grey forest type. *'Polis´ka 90'* and *'Stolychna'* were selected at NSC Institute of Agriculture of NAASU. *'Stolychna'* was established by individual selection of hybrid combination of crossing cultivars *'Polis'ka 92'/'Kolosysta'/'Polis'ka 90'*. *'Stolychna'* could be more tolerant to fungal pathogens than *'Polis´ka 90'*.

The 0.1 mM oxalic acid solution was sprayed on wheat plants in booting phase. After this plants were inoculated by *S. tritici*. Oxalic acid has three chemical natures: it is as proton and electron source, and a strong metal chelator, despite its simple chemical formula of $(COOH)_2$. Due to its unique multiple chemical natures, it has been receiving much attention for its various ecological qualities, such as: (i) bioremediation of a wide variety of organic pollutants with lignin biodegradation systems; (ii) inactivation of copper-containing wood preservatives by wood-rotting fungi; (iii) detoxification of aluminum toxicity in Al-resistant buckwheat; (iv) crop damage caused by oxalic acid-producing phytopathogens; (v) the biofertilizer effect of ectomycorrhizal fungi; and (vi) being an electron source for nitrogen fixation in symbiotic rhizobia in a legume plant (Lane 2002).

The degree of infection was estimated on Saari and Prescott scale in milky ripeness phase of grain development. The yield structure analysis included plant height, grain quantity in ear, flag leaf and ear length. The experiments were repeated 3 times and data were statistically analyzed by ANOVA.

Results and discussion

The fungus causes pale grey to dark brown blotches on the leaves, and, in a lesser extent, stems and ears (Fig. 1 B; Fig. 2 B; Fig. 3 B). When the disease is severe, entire leaves may be affected by disease lesions. The diagnostic feature of Septoria tritici blotch is the presence of black fruiting bodies (pycnidia) within the blotches. These tiny black spots give the blotches a characteristic speckled shape (Бабаянц 1988).

Saari and Prescott scale has 9 points. It is shown that oxalic acid decreased about 1-2 points of this scale in both winter wheat varieties. The wheat leaves treated by oxalic acid have decreasing about 10-15% of infected square compared with untreated plants (Fig. 1 C; Fig. 2 C; Fig. 3 C). Infected plants have more fragile stem than control plants.

Effect of elicitors – oxalic acid – is increased quantity of grains per ear and plant height in cultivar *'Stolychna'*. In the more sensitive variety *'Polis´ka 90'* treatment by oxalic acid stimulated growth of flag leaf in infected plants. Oxalic acid stimulated growth and development of winter wheat cultivars *'Polis´ka 90'* and *'Stolychna'*.

Most fungus infection observed in endospermal part of grains. The development of fungal mycelium destroyed envelope of the endosperm, the hyphae of the fungus penetrated into the endosperm tissue to a considerable depth and caused the formation of necrotic spots. In plants that were infected with the fungus in booting phase and treated by oxalic acid the pathogen lesions were reduced (Fig. 1). In grain endosperm the small dark spots were detected, what indicates poor development of fungal mycorrhizae and increased resistance to penetration hyphae of the fungus to the grain tissues of the wheat.

Oxalic acid treatment stimulated stem growth under conditions of defeat fungal pathogen agent because it can stimulate the antioxidant defense system as it was shown in our previous investigation (Жук і Дмитрієв 2013) The average length of wheat leaves, height, and grain quantity were decreased under Septoria tritici blotch but treatment by oxalic acid increased these morphometric parameters. This fact suggests that oxalic acid has a good potential for practical application as an elicitor. The data obtained suggest that exogenic oxalic acid decreased the degree of leaf

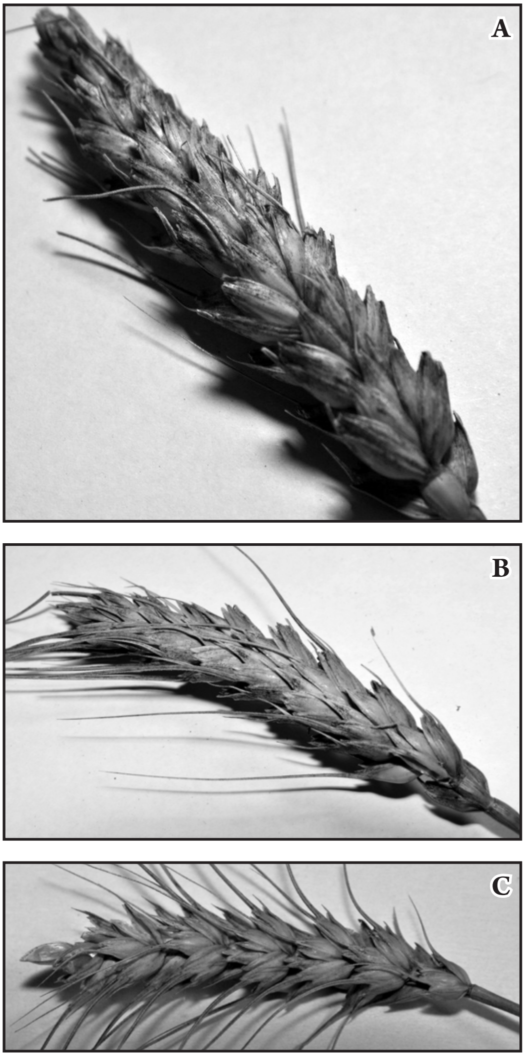

Fig. 2. The ears of winter wheat cultivar *'Polis´ka 90'*: **A** – control; **B** – infected by winter wheat leaf blotch; **C** – infected by winter wheat leaf blotch under oxalic acid treatment.

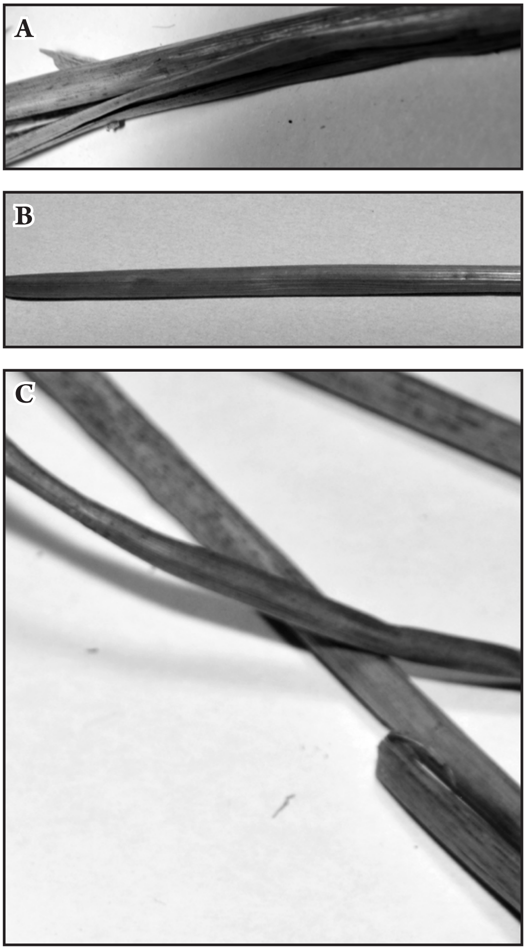

Fig. 2. The leaves of winter wheat cultivar *'Polis´ka 90'*: **A** – control; **B** – infected by winter wheat leaf blotch; **C** – infected by winter wheat leaf blotch under oxalic acid treatment.

damage and increased last leaf length, as well as grain quantity.

Conclusions

1. It is shown that oxalic acid is effective for decreasing growth and development of fungal pathogen *S. tritici* under field conditions and has a good potential for practical application as an elicitor.
2. The treatment by oxalic acid decreases the infected square of flag leaves.
3. Oxalic acid reduces the development of fungal mycorrhizae, and increases resistance to penetration of fungal hyphae into the grain tissues of *T. aestivum*.

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