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ВЛИЯНИЕ ЭКЗОМЕТАБОЛИТОВ ФИТОПАТОГЕНОВ Р. *FUSARIUM* НА МОРФОФИЗИОЛОГИЧЕСКИЕ ОСОБЕННОСТИ КАЛЛУСНЫХ КУЛЬТУР ИЗОГЕННЫХ ПО ГЕНАМ *VRN* ЛИНИЙ ПШЕНИЦЫ

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В культуре *in vitro*, изучали влияние культуральных фильтратов микромицетов *Fusarium oxysporum* и *Fusarium moniliforme* на рост каллусов и активность пероксидазы изогенных по генам контроля темпов развития (*VRN*) линий пшеницы *Triticum aestivum* L. В результате проведенных исследований установлено, что экзометаболиты фитопатогенов, оказывают противоположные эффекты на цитологические параметры (число и длина каллусных клеток), рост каллусных культур (ПИ) и ферментативную активность пероксидазы у изолиний, различающихся темпами развития в условиях *in vivo*. Культуральный фильтрат *F. oxysporum* оказывает больший токсический эффект на рост каллусных культур по сравнению с экзометаболитами *F. moniliforme*. Предполагается, что генетическая система контроля темпов развития *Triticum aestivum* L. *in vivo* опосредованно детерминирует формирование устойчивости к биотическому стрессу в условиях *in vitro*.

Ключевые слова: *Triticum aestivum* L., *Fusarium oxysporum*, *Fusarium moniliforme*, гены *VRN*, яровизация, NILs, каллусная культура, ростовой индекс, устойчивость к фитопатогенам.

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INFLUENCE OF EXOMETABOLITES PHYTOPATHOGENS G. *FUSARIUM* ON MORPHO-PHYSIOLOGICAL CHARACTERISTICS OF CALLUS CULTURES OF ISOGENIC BY GENES *VRN* WHEAT LINES

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Wheat is the most valuable food crop taking the first place in cereal balance of Ukraine [Morgun and al., 2010]. Its efficiency depends on the realization of genetically laid characteristics and influence of environmental conditions reacting at each stage of plant ontogenesis [Morgun and al., 2010]. It is known, that duration of ontogenesis, type of development (spring/winter) and speed of development in soft wheat *Triticum aestivum* L. determined by the system of *VRN* genes [Stelmach and al., 2000; Cockram J. and al., 2007]. One of the factors limited the productivity of soft wheat is affection by vascular diseases induced by different types of micromycete of g. *Fusarium* [Grutcyk, 2013]. *In vitro* is a modern model system in phytobiological research and in present days is widely used in cell selection for receiving

stable to diseases plant varieties [Bavol and al., 2009; Kornya, 2011]. Thus, in forming of resistance to plant pathogens, age, plant ontogenesis phase put through biotic stress are of the essence, it is interesting to learn predetermination by genes of wheat development rates in forming of biological mechanisms of resistance to biotic stresses.

The aim of our work is to research the influence of exometabolites phytopathogens of genus *Fusarium* on callus cultures isogenic wheat lines differ by development rates. The objects of research were almost isogenic by genes *VRN* lines (NILs) of soft wheat *Triticum aestivum* L., Mironovskaya 808 sort and phytopathogenic micromycetes *Fusarium oxysporum* and *Fusarium moniliforme* (micromycet culture collection of Physiology and Biochemistry of Plant and Microorganisms

Department V. N. Karazin Kharkiv National University). Primary callus cultures of isogenic lines were obtained using in the capacity of explants mature embryo. The cultivation was conducted in nutrient medium Myrasige and Skuga (MS) containing growth stimulant – 2,4-D (2 mg/l) in thermostat with temperature 26°C in the darkness. Phytopathogens exometabolites influence g. *Fusarium* was researched by adding cultural filtrate of micromycetes into nutrient medium MS in the ratio 1:20. The cultivation of callus cultures of isogenic lines was conducted during four weeks analyzing growing indicators (callus areas, growing index, length and number of cells of callus tissues). On the finishing of cultivation, we conducted peroxidase activity determination – the main component in a enzymatic antioxidant plant system.

The results of our experiments showed that in conditions of *in vitro* cultures exometabolites phytopathogens *F. oxysporum*

insignificantly and *F. moniliforme* considerably stimulate the growth of callus cultures of fast-developing isogenic lines *VRN Ala* and *VRN D1a* (table). Growing index of callus cultures *VRN Bla* and sort (all *vrn* genes are recessive) under the action of exometabolites *F. oxysporum* is reducing and under the action of *F. moniliforme*, on the contrary, is growing. Application of cultural filtrate into the medium of callus cultivation influence on their cytological characteristics. Isolines differ by development rates differently react on exometabolites plant pathogens. In fast-developing, in conditions *in vivo*, isolines *VRN Ala* and *VRN D1a* in callus culture under the influence of phytopathogens the number of cells is reducing, but their length is growing. In slowly-developing isolines *Vrn Bla* and sort, on the contrary, the number of cells is growing, but its size is reducing.

Table. The influence of exometabolites g. *Fusarium* on the growth of callus cultures of isogenic by genes *VRN* wheat lines

Isoline	Variant	Growing index, %	Number of cells, N×10 ⁶ /g	Length of cells, mcm	Peroxidase activity, y.e
VRN A1a	Control	21.2	12.5	6.0	2.54
	<i>F. oxysporum</i>	26.8	9.9	10.2	4.08
	<i>F.moniliforme</i>	31.5	9.1	9.5	3.47
VRN B1a	control	18.2	5.9	13.3	8.83
	<i>F. oxysporum</i>	11.4	7.7	9.3	2.63
	<i>F.moniliforme</i>	21.1	12.7	12.5	2.89
VRN D1a	control	15.3	6.0	6.1	1.05
	<i>F. oxysporum</i>	24.3	5.2	6.7	2.23
	<i>F.moniliforme</i>	37.6	5.9	8.4	1.87
Sort M-808 (all the <i>vrn</i> genes are recessive)	control	18.9	5.6	9.3	4.16
	<i>F. oxysporum</i>	16.3	7.2	6.3	1.95
	<i>F.moniliforme</i>	25.9	8.1	7.1	2.57
HCP _{0,05}		2.4	1.2	1.9	0.9

It is necessary to note, that exometabolites *F. oxysporum* are more phytotoxic, which is seen in changing of all the morpho-physiological callus cultures indicators in comparison to *F. moniliforme*. Among researched isogenic lines of wheat, the most resistant to pathogens by all indicators is isolate *VRN Ala*, characterized by fast rates of growing plant conditions

of *in vivo*, and the less resistant – slowly-developing isolate *VRN Bla*. The received results allow us to suppose that *VRN* genes, determining the rates of development in conditions *in vivo* indirectly in participate in forming of resistance to exometabolites phytopathogens g. *Fusarium* in conditions *in vitro*.

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In the culture *in vitro*, we studied the influence of cultural micromycetes filtrates of *Fusarium oxysporum* and *Fusarium moniliforme* on callus growth and peroxidase activity of development rates (*VRN*) of isogenic by genes *Triticum aestivum* L. wheat lines. As a result, of the conducted researches it is established that exometabolites phytopathogens render opposite effects on cytological parameters (number and length of callus cells), the growth of callus cultures (GI) and peroxidase fermentative activity at the isolines differing with development rates in the conditions of *in vivo*. The cultural filtrate of *F. oxysporum* renders bigger toxic effect on the growth of callus cultures in comparison with *F. moniliforme* exometabolites. It is supposed that the genetic monitoring system of *Triticum aestivum* L. wheat development rates in the conditions of *in vivo* indirectly determines the formation of the resistance to biotic stress in the conditions of *in vitro*.

Keywords: *Triticum aestivum* L.; *Fusarium oxysporum*; *Fusarium moniliforme*; *VRN* genes; vernalization; NILs; callus culture; growth index; resistance to phytopathogens.