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THE HERITABILITY OF TOLERANCE TO *Alternaria dauci* AND *Fusarium oxysporum* IN F₁-HYBRIDS OF CARROT

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Abstract

Heritability the tolerance to *Alternaria dauci* and *Fusarium oxysporum* in prospective F₁-hybrids of carrot has been discussed in the paper. As QTLs isn't reliable enough for the determination of tolerance heritability in F₁-hybrid progenies, authors used traditional methods of inoculation applying step-by-step at every stage of plant's development. Carrot lines have been evaluated carefully at different types of infection backgrounds, tolerant lines and lines with weak sensitivity have been selected for crossings. F₁-hybrids have been obtained and evaluated at the provocative infection background. Using dispersion analysis, the authors show, that tolerance to *Alternaria dauci* and *Fusarium oxysporum* has been inherited by the paternal type.

Key words: carrot, resistance, tolerance, heritability, *Alternaria dauci*, *Fusarium oxysporum*

Introduction

Alternaria dauci, *Alternaria radicina* and *Fusarium oxysporum* are the most spread and damaging fungal pathogens affecting carrots in Moscow region, Russian Federation, and in the world. *Alternaria dauci* causes leaf blight and premature death of leaves, which makes many difficulties for using mechanization tools during harvest roots. Besides, the content of carotene in roots decreases on 24%, and the content of sugars decreases on 31%, so the quality of production is lowered. *A. radicina* causes rots 50-70% of carrot roots during storage. *Fusarium oxysporum* causes the tracheomycosis wilt, retardation growth of the plant, root and stem rots, "black leg" of seedlings. Obtaining new varieties and hybrids of carrot with resistance to these pathogens is complicated by the fact, that carrot resistance to fungal diseases has polygenic control. Eleven quantitative trait loci (QTLs) in two genetic backgrounds of carrot, resistant to *Alternaria dauci* were identified by Le Clerc V. with collaborators in 2015 (1). But only 4 of them were constant across years of studies, seven were detected within a single year. Authors concluded: instability of QTLs during years could be conditioned with changing of environmental characteristics. So, there are 2 main questions for carrot breeders in that situation:

- How resistance/tolerance to each pathogen will be inherited in F₁ –progenies?
- How to select parental pairs in such case?

The goal of our study is to determine the heritability of carrot tolerance/field resistance to *Alternaria dauci* and *Fusarium oxysporum* in prospective F₁- hybrids.

Materials and methods

Objects of studies were: 7 lines of carrot, which were obtained from carrot populations with different type of roots; 7 F₁ –hybrids of carrot, which were obtained from crossings of above lines; mycelium of studied pathogens with spores for artificial inoculation.

Methods of studies. We used PCR for determination accessory of local species the pathogens of *Fusarium* and *Alternaria* genus. As QTLs isn't reliable enough for the determination of carrot resistance, we used traditional methods of inoculation for that purpose. But they were applied step-by-step at every stage of plant development for the modeling every type of epidemiologic situation.

1. Seedlings growing at the filtrate of *Fusarium oxysporum* cultural liquid and selection the resistant ones.
2. Spray growing carrot plants of the first year with spore solution of *Alternaria dauci*.
3. Carried in the soil mycelium of *Fusarium oxysporum* and *Alternaria radicina* with spores
4. Spray of growing plants of the second year with spore solution of *Alternaria dauci*.

Then we evaluated resistance of plants by known scales have been developed by us earlier (2) and selected tolerant plants. Data of evaluation have been calculated with dispersion and correlation analyses (3).

Results and discussions

1. First of all, with help of PCR we determined local species fungi of *Fusarium* and *Alternaria* genus, and confirmed their accessory to *Fusarium oxysporum*, *Alternaria radicina* and *Alternaria dauci* (Fig.1, 2, 3).

Fig.1 Electrophoresis of PCR-products with primer FuzOxF/FuzOxR for the determination of *Fusarium oxysporum*. Marker fragment about of 500 b.p. is identified at 6p, 7p-samples.

Fig.2. Electrophoresis of PCR-products with primer for the determination of *A. radicina*.

Fig.3. Electrophoresis of PCR-products with primers for the determination of *Alternaria dauci*.

2. Than we evaluated line resistance with help of artificial infection applied step-by step (Table1), and selected of maternal and paternal forms for crossings. Line # 1268 with tolerance to group of pathogens we used as paternal form, and other lines with weak sensitivity to group of pathogens we used as maternal forms. F₁ - hybrids have been obtained resulted in crossings.

Table 1 . Line resistance/tolerance to *Fusarium oxysporum*, *Alternaria dauci*, *Alternaria radicina*. FSVC. 2011-2012

Lines	Groups of resistance at the year of vegetation								
	2011 (first year)					2012 (second year)			
	Seedlings			Leaves		Roots		Leaves	
	<i>Fusarium oxysporum</i>			<i>Alternaria dauci</i>		<i>F. oxysporum</i> , <i>A. radicina</i>		<i>Alternaria dauci</i>	
	FCL*	AIB	PIB	PIB	NIB	PIB	NIB	PIB	NIB
1268	T	T	R**	T	T	T	T	R	R
1238B	WS	WS	T	WS	WS	T	T	T	T
661B	WS	WS	T	T	WS	WS	T	T	WS
22	T	WS	T	T	T	T	T	WS	T
K45	WS	WS	T	WS	T	T	T	WS	T
MC1-1	WS	WS	T	T	T	WS	T	T	T
45	T	WS	T	T	WS	T	T	WS	T

*FCL-filtrate of cultural liquid, AIB – artificial infection background, PIB-provocative infection background, NIB – natural infection background;**R-resistant, T-tolerant, WS-weak sensitive

3. Seven F₁-hybrids were evaluated at the artificial (provocative) infection background of *Alternaria dauci* (table2). 3 of them were tolerant to *Alternaria dauci* at the artificial infection background: K45x1268, MC 1-1x1268, K45x225B. Line # 1268 was paternal form from 2 of them.

Table 2. Sensitivity to *Alternaria dauci* the F₁-hybrids of carrot. FSVc. 2014-2019

F ₁ -hybrids	Degree of leaves affection, middle mark						Σ_v	□	Deviation from St _{tol.}
	2014	2015	2016	2017	2018	2019			
22x1268	1,4	1,4	1,3	1,1	1,1	0,9	7,2	1,20	0,35
K45x1238B	1,2	1,5	1,5	1,4	1,5	1,4	8,5	1,42	0,57
22x661B	1,2	1,7	1,6	1,2	1,1	1,1	7,9	1,32	0,47
MC1-1x1268	1,6	1,4	1,3	1,1	0,8	0,8	7,0	1,17	0,32
K45x1268	0,9	0,8	0,9	0,7	0,9	0,9	5,1	0,85	St_{tol.}
K45x225B	1,2	1,4	1,3	1,1	1,1	1,0	7,1	1,18	0,33
45x225B	1,5	1,3	1,4	1,5	1,2	1,1	8,0	1,33	0,48
Σ_p	9,0	9,5	9,3	8,1	7,7	7,2	50,8	LSD ₀₅	0,34

* St_{tol.} – Standard of tolerance

These 7 hybrid combinations were evaluated at the artificial (provocative) infection background of *Fusarium oxysporum* (table3). 4 of them were selected as tolerant to *Fusarium oxysporum* at the artificial infection background: 22x1268, MC1-1x1268, K45x1268, K45x225B. Line # 1268 was paternal form from 3 of them.

4. Then we studied the heritability of tolerance to *Alternaria dauci* and *Fusarium oxysporum* in F₁-hybrids of carrot with help of the heritability coefficient. The heritability coefficient can be calculated with help of correlation and dispersion analysis (3). First of all, we determined the role of maternal lines in the formation of tolerance to *A. dauci* in F₁-hybrids, using 22 co-variation pairs in correlation analysis. The correlation coefficient was 0,07 and the correlation wasn't significant. So, the tolerance to *A. dauci* isn't inherited by maternal type. It may be inherited by paternal type. Second, we determined the heritability tolerance to *F. oxysporum*. The variability of maternal line tolerance didn't correlate with the variability of F₁-hybrids tolerance. The correlation coefficient was 0,23. Correlation wasn't significant. Tolerance to *F. oxysporum* isn't inherited by maternal type. It may be inherited by paternal type.

Table 3. Sensitivity to *Fusarium oxysporum* the F₁-hybrids of carrot. FSVc. 2014-2019

F ₁ -hybrids	Degree of leaves affection, middle mark						Σ_v	□	Deviation from St _{tol.}
	2014	2015	2016	2017	2018	2019			
22x1268	1,5	1,4	1,2	1,0	1,0	0,9	7,0	1,16	0,21
K45x1238B	1,5	1,5	1,4	1,2	1,1	1,0	7,7	1,28	0,33
22x661B	1,4	1,3	1,2	1,2	1,0	1,0	7,1	1,18	0,23
MC1-1x1268	1,5	1,3	1,3	1,1	0,9	0,8	6,9	1,15	0,20
K45x1268	1,1	1,1	1,0	0,9	0,8	0,8	5,7	0,95	St_{tol.}
K45x225B	1,2	1,2	1,3	1,1	1,0	0,8	6,6	1,10	0,15
45x225B	1,5	1,4	1,3	1,4	1,2	1,1	7,9	1,32	0,37
Σ_p	9,7	9,2	8,7	7,9	7,0	6,4	48,9	LSD ₀₅	0,22

* St_{tol.} – Standard of tolerance

5. The role of paternal lines in the formation of tolerance to *A. dauci* and to *F. oxysporum* we determined with dispersion analysis, using F₁-hybrids have been obtained resulted in crossings of 1 maternal line and 3 paternal lines (tables 4-5, 6-7).

F_{fact.} > F₀₅: inclusion of paternal forms in the tolerance appearance in F₁-hybrids is significant: the tolerance to *A. dauci* can be inherited by the paternal type. Genotypic and phenotypic variability $s_g^2 = 0,0785$; $s_{ph}^2 = 0,0925$; $h^2 = 0,85$ or 85%. So, selection of paternal forms by this exponent will be effective.

Table 4. Analysis the heritability of tolerance to *Alternaria dauci* in F₁-hybrids of carrot. FSVc. 2014-2019

Maternal line	Paternal line	Degree of leaves affection, middle mark						Σ_v	□
		2014	2015	2016	2017	2018	2019		
K-45	1238B	1,2	1,5	1,5	1,4	1,5	1,4	8,5	1,42
K-45	1268	0,9	0,8	0,9	0,7	0,9	0,9	5,1	0,85
K-45	225B	1,2	1,4	1,3	1,1	1,1	1,0	7,1	1,18
Σ_p		3,3	3,7	3,7	3,2	3,5	3,3	20,7	-

Table 5. Results of dispersion analysis

Dispersion (name)	Dispersion (number)	Degrees of freedom	The middle square	F _{fact}	F ₀₅
Common	1,18	17	-	-	-
Repetitions	0,07	5	-	-	-
Father lines	0,97	2	0,485	34,64	4,10
Remainder (mistake)	0,14	10	0,014	-	-

Table 6. Analysis the heritability of tolerance to *Fusarium oxysporum* in F₁- hybrids of carrot. FSVC. 2014-2019

Maternal line	Paternal line	Degree of leaves affection, middle mark							Σ _v
		2014	2015	2016	2017	2018	2019		
K-45	1238B	1,5	1,5	1,4	1,2	1,1	1,0	7,7	1,28
K-45	1268	1,1	1,1	1,0	0,9	0,8	0,8	5,7	0,95
K-45	225B	1,2	1,2	1,3	1,1	1,0	0,8	6,6	1,1
Σ _p		3,8	3,8	3,7	3,2	2,9	2,6	20,0	-

Table7. Results of dispersion analysis

Dispersion (name)	Dispersion (number)	Degrees of freedom	The middle square	F _{fact.}	F ₀₅
Common	0,82	17	-	-	-
Repetitions	0,44	5	-	-	-
Father lines	0,34	2	0,17	42,5	4,10
Remainder (mistake)	0,04	10	0,004	-	-

F_{fact.} > F₀₅: inclusion of paternal forms in the tolerance appearance in F₁-hybrids is significant: the tolerance to *F. oxysporum* can be inherited by the paternal type. Genotypic and phenotypic variability $s_g^2 = 0,0277$; $s_{ph}^2 = 0,0317$; $h^2 = 0,87$ or 87%. So, selection of paternal forms by this exponent will be effective.

Resulted in the studies we received the increasing % of resistant plants to *Alternaria dauci* and to *Fusarium oxysporum* in F₁-hybrid populations of carrot evaluated at the provocative infection backgrounds for 3 F₁-hybrid combinations: 22x1268, MC1-1x1268, K45x1268. This increasing wasn't harsh, it was flowing, but stable for 9 years.

Conclusions

1. Isolates of fungal pathogens affected carrot in Moscow region accessed to *Fusarium oxysporum*, *Alternaria radicina* and *Alternaria dauci*.
2. The tolerance to *Alternaria dauci* and the tolerance to *Fusarium oxysporum* can be inherited in F₁-hybrids of carrot by paternal type.

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