PATHOLOGICAL CHANGES IN SUNFLOWER INFECTED BY *FUSARIUM* SPECIES OF DIFFERENT PATHOGENICITY

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Abstract

Lately, the widespread disease of many crops caused by fungi of the genus *Fusarium* became a frequent occurrence in sunflower in Russia, including the Krasnodar region. The most pathogenic and phytotoxic strains of monospore cultures of two different *Fusarium* species spreading on sunflower: *F. oxysporum* var. *orthoceras* (average pathogenicity) and *F. sporotrichiella* var. *sporotrichioides* (high pathogenicity) have been isolated. Experimental modeling of host-pathogen biosystems of vegetative sunflower plants (two cultivars and two hybrids) with these strains of each fungal species was carried out. It was revealed that the fungal species, differing in pathogenicity, equally significantly decrease plant height, head diameter and mass of 1000 seeds of susceptible sunflower plants, both cultivars and hybrids, that may be proof of nutrition structure likeness of these fungal species. Considerable differences in yield decrease of various infected sunflower genotypes were also observed, and this fact could be the consequence of their diverse resistance. Significant oil content decrease was selective for each system depending on host genotype; this could be connected both with different fungal pathogenicity and with their unequal adaptation to various sunflower genotypes. As a whole, the disease has harmful effects for sunflower plants.

Introduction

In the last decade of the last century a disease of sunflower caused by fungi of the genus *Fusarium* spread in fields of the Russian Federation, including the Krasnodar region. Thus, there arose a real necessity to carry out sunflower breeding for resistance to the fungi of these species.

The disease, caused by *F. moniliforme* and *F. oxysporum* in India and North America was described in the 1970s (Orellana, 1971; Ghodajkar et al., 1976; Bhargava et al., 1978). During the same period of time in Texas there were cases of from 40% up to 90% of yield losses due to the head infection by *F. moniliforme* (Orellana, 1971). In the beginning of the 1980s in Italy, *F. tabacinum* species, which is found on sunflower only in this country, was also described (Zazzerini and Tosi, 1987). The well-known monograph of Bilai (1977) also contains records on sunflower head infection by *F. moniliforme* in the former USSR territory. But already in the middle of the 1990s in the Belgorod and Voronezh regions, more species: *F. sporotrichiella* var. *sporotrichioides, F. sambucinum, F. avenaceum*, and *F. solani* were isolated from infected sunflower seeds and the problem of sunflower infection by *Fusarium* became obvious (Yakutkin, 1995).

Purposive investigations of sunflower infection by *Fusarium* causal agents were started in VNIIMK in 1999. Twelve varieties of *Fusarium* ssp. isolated from infected sunflower plants in the Krasnodar region have been identified: *F. oxysporum* (Schlecht). emend. Snyd. and Hans, *F. oxysporum* var. *orthoceras* (App. and Wr.), *F. sporotrichiella* var. *poae* (Pk.) Wr. emend. Bilai, *F. sporotrichiella* var. *tricinctum* (Cda.) Bilai, *F. sporotrichiella* var. *sporotrichioides* (Sherb.) Bilai, *F. semitectum* Berk. and Rav., *F. gibbosum* App. and Wr. emend. Bilai, *F. moniliforme* (Scheldon), *F. solani* (Mart.) App. and Wr., *F. solani* var. *argillaceum* (Fr.) Bilai, *F. javanicum* Koord, and *F. heterosporum* Nees (Maslienko and Muradosilova, 2000; Saukova, 2001; Antonova et al., 2002a; Antonova et al., 2002b). It has been proven that the most comon isolate on sunflower is *F. oxysporum* var. *orthoceras* (about 60% of all isolates). Sunflower infection by *F. sporotrichiella* varieties in some areas raises serious concerns because of its toxicity both for humans and animals. Thus, breeding work for this varietal resistance should be carried without hesitation.

The aim of this work was to investigate harmful effects of the disease by experimental injection into sunflower of the most pathogenic isolates, *F. oxysporum* var. *orthoceras* and *F. sporotrichiella* var. *sporotrichioides*. Actually, these investigations explain the need for the creation of high levels of resistance in breeding lines.

Materials and Methods

Sunflower varieties SPK, VNIIMK 8883 and hybrids Kubansky 930, Kubansky 939 were cultivated on four-row (26 plants in a row) randomised plots, repeated three times. The sowing was carried out by square-cluster planting with five seeds in a hole, reserving two plants at thinning. The square of plots was 25.5 m sq. (104 plants). Forced injection was carried out separately with two fungus varieties: *F. oxysporum* var. *orthoceras* (average pathogenicity) and *F. sporotrichiella* var. *sporotrichioides* (high pathogenicity).

Sunflower plants in the vegetative stage were infected twice by the injection of a spore suspension into the hypocotyls at the 4-5 pairs of true leaves stage and in stems during the budding stage. On experimental plots all plants were infected. The plants of control plots were given a distilled water injection instead of a spore suspension.

A spore suspension of each fungus was prepared in the following way: spores of 14-day monospore colonies, cultivated on agar at room temperature and natural lighting, were washed off with a brush in distilled water. Concentration of spores in suspension was 1.2×10^6–1.2×10^7 spores/ml. Each injection contained 0.25 ml of suspension.

Plants of each plot with evident disease symptoms were marked. In the ripening stage plant height and head diameter were measured. Also, absolute dry mass of 1000 seeds and average yield per plant were estimated after individual threshing of heads. Oil content was defined by the nuclear magnetic resonance method using an AMV 1006 M analyzer. Results of all experiments were analyzed by using the single-factor dispersion analysis (Dospechov, 1985).

Results and Discussion

It was observed that after double inoculation of the hypocotyl and stem, the plants were equally resistant and susceptible to infection. Dried and lignified spots from 0.2 to 1.5 cm in diameter remained at the injection points of resistant plants. Susceptible plants were characterized by stem rot, and black circular necrosis near the hypocotyl base in the first injection point; the second injection point could be easily seen, proving unrestricted fungus development. Apart from resistant and highly susceptible plants, a group of tolerant plants could be distinguished. They had no evident infection traits, only a slight tint of beige on the stem. The presence of the fungus inside was later proved by subsequent fungus reisolation.

Susceptible plants of all investigated genotypes with evident infection traits when infected by each fungus species, in comparison with the control inoculated by distilled water, had a significant decrease in height and head diameter, mass of 1000 seeds and yield (Tables 1 and 2).

Table 1. Pathological changes of sunflower after artificial infection by *F. oxysporum* var. *orthoceras* VNIIMK, 2002.

Varieties, hybrids	Variants*	Plant height (cm)	Head diameter (cm)	Mass of 1000 seeds (g)	Oil content (%)	Yield (g/plant)
VNIIMK	1	174	14	38	48,2	41,9
8883	2	196	18	51	53,3	57,6
	L.S.D. at 0.05	5,2	1,2	3,8	3,5	10,9
	1	180	15	55	45,1	46,1
SPK	2	216	19	82	48,5	91,7
	L.S.D. at 0.05	11,8	1,9	9,2	3,5	13,1
Kubansky	1	171	14	41	48,8	39,9
930	2	190	21	57	51,2	104,1
	L.S.D. at 0.05	6,9	4,4	6,2	2,9	24,9
Kubansky	1	168	16	47	49,6	43,7
939	2	181	21	58	51,4	60,0
	L.S.D. at 0.05	6,1	2,9	3,9	1,7	5,3
* -1 – infected plants; 2- healthy plants						

Though *F. oxysporum* var. *orthoceras* species is less pathogenic than the *F. sporotrichiella* var. *sporotrichioides* species, each genotype had almost the same decrease in plant height and head diameter in both cases. Height decrease in hybrid Kubansky 939 was 10-12 cm and variety SPK 35-36 cm. Height decrease of the other two genotypes was within 20-22 cm. A decrease in both varieties head diameter was 5-7 cm.

Both fungi caused almost the same decrease in mass of 1000 seeds in each variety (12-13 g and 22-25 g, respectively) and hybrid Kubansky 930 (16-18 g). However, infection of hybrid Kubansky 939 by the more pathogenic fungus caused significantly greater (17 g) decrease in mass of 1000 seeds in comparison with infection by the less pathogenic fungus (11 g).

Infection by each fungus species caused a different decrease in average yield of one plant. Variety VNIIMK 8883 reacted by a yield decrease of 20-24%, hybrid Kubansky 939 by 27-31%; whereas variety SPK and hybrid Kubansky 930 turned out to be more susceptible (50-62 % and 61,5-62% correspondingly). But, in spite of the different pathogenicity both fungi caused the same high yield decrease of hybrid Kubansky 930.

Table 2. Pathological changes of sunflower after artificial infection by *Fusarium sporotrichiella var. sporotrichioides* VNIIMK 2002 .

Varieties, hybrids	Variants*	Plant height (cm)	Head diameter (cm)	Mass of 1000 seeds (g)	Oil content (%)	Yield (g/plant)
VNIIMK	1	173	14	39	49,6	44,2
8883	2	196	18	51	53,3	57,6
	L.S.D. at 0.05	5,5	1,9	3,5	4,4	6,4
	1	181	15	60	44,8	34,8
SPK	2	216	19	82	48,5	91,7
	L.S.D. at 0.05	14,2	2,5	9,6	3,6	10,2
Kubansky	1	170	14	39	48,9	38,6
930	2	190	21	57	51,2	104,1
	L.S.D. at 0.05	5,3	5,4	5,4	1,9	23,8
Kubansky	1	171	14	41	48,4	41,1
939	2	181	21	58	51,4	60,0
	L.S.D. at 0.05	5,1	2,8	4,9	2,6	6,1
* -1 – infected plants; 2- healthy plants						

Both fungi caused oil content of seeds to decrease in some genotypes. *Fusarium oxysporum* var. *orthoceras* caused significant oil content decrease in variety VNIIMK 8883 (by 5.1 %) and hybrid Kubansky 939 (by 1.8 %). The more pathogenic fungus *F. sporotrichiella* var. *sporotrichioides* was the cause of a more considerable oil content decrease in both hybrids (2.3 % and 3.0 %) and variety SPK (3.7 %), whereas VNIIMK 8883 had a nonsignificant decrease. Very likely, these distinctions are connected with different fungal pathogenicity, because variety SPK and hybrid Kubansky 930, according to oil content of their seeds were more resistant to the less pathogenic fungus, while the more pathogenic fungus caused an oil content decrease in these plants. Inverse dependence of variety VNIIMK 8883 can be explained by the fact that the second fungus has not spread enough and is insufficiently adapted to this genotype, whereas *F. oxysporum* var. *orthoceras* species prevails in *Fusarium* populations able to infect sunflower.

Conclusions

Thus, in host-pathogen experimental systems, sunflower with two *Fusarium* species different in their pathogenicity have similar pathological aftereffects (for susceptible plants) decreasing plant height, head diameter, and mass of 1000 seeds. Significant differences in yield decrease of infected plants of various genotypes are the proof of different resistance degrees of the latter. Considerable oil content decrease is selective for each system depending on genotype, and this can be attributed both to different fungal pathogenicity, and their unequal adaptivity to various sunflower genotypes. As a whole, the disease has harmful effects for sunflower plants.

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References

Antonova, T.S., Maslienko, L.V., Muradosilova, N.V., and Saukova, S.L. 2002a. Fungi from genus *Fusarium* are on sunflower in Krasnodar region and their pathogenicity. Modern Mycology in Russia. The 1st Conference of mycologists. Abstracts. Moscow, 171.

Antonova, T.S, Araslanova, N.M., and Caukova, S.L. 2002b. Prevalence of *Fusarium* spp. on sunflower in Krasnodar region. Reports of Russian Academy of Agricultural Sciences. 3:6-8.

Bhargava, S.N., Shukla, D.N. and N. Singh. 1978. Wilt of sunflower caused by *Fusarium moniliforme*. Indian Phytopathologist. 31: 382.

Bilai, V.I. 1977. *Fusariums*. Kiev: "Naukova Dumka ," 442.

Dospechov, B.A. 1985. Methods of field experiment. Moscow, "Agropromizdat," 351.

Ghodajkar B.N., Ganacharya, N.M., Bindu, K.J. and Jadhav, V.T. 1976. Sunflower wilt, a new disease from India. Current Science. 45:844.

Maslienko, L.V., and Muradosilova, N.V. 2000. Specific composition of *Fusarium* spp. on sunflower. Scientific-technical bulletin of VNIIMK. Krasnodar. 123:25-31.

Orellana, R.G. 1971. *Fusarium* wilt of sunflower, *Helianthus annuus*: First report. Plant Diseases Report. 55:1124-1125.

Saukova, S.L. 2001. Fungi varieties of *Fusarium* spp. on sunflower and seedlings infection by different doses of inoculum. ScientificTechnical Bulletin of VNIIMK. Krasnodar. 124:173-175.

Yakutkin, V.I. 1995. Plant protection in conditions of agro-industrial complex reforming: economic, efficiency, protection (abstracts). St. Petersburg, 108.

Zazzerini, A. and Tosi. L. 1987. New sunflower disease caused by *Fusarium tabacinum*. Plant Diseases. 71:1043-1044.