RESEARCH CONCERNING RESISTANCE TESTS OF SUNFLOWER AGAINST THE PATHOGEN DIAPORTHE/PHOMOPSIS HELIANTHI (MUNT. CVET. ET AL.)

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Abstract

A number of 1560 sunflower genotypes have been tested in 1989-1993 under natural infection conditions at Fundulea, of which 450 being trialled both under natural and artificial infection with *Diaporthe/Phomopsis helianthi* (Munt. Cvét. et al.). A high resistance level has been recorded in lines with cytoplasmic androsterility AC-1 (LC-1004), AC-2 (LC-1020), AC-3 and AC-4, as well as in the restorer lines RF-1, RF-2 and RF-3. A resistant hybrid can be obtained if both parents are resistant to Phomopsis. Recurrent selection is the best way to improve resistance to Phomopsis. A positive correlation has been found between the attack level and yield loss by natural and artificial infection with *Diaporthe/Phomopsis helianthi* (Munt. Cvét. et al.).

Key-words : sunflower stem canker - *Diaporthe/Phomopsis helianthi* (Munt. Cvét. et al.), natural infection, artificial infection, additivity, recurrent selection.

Introduction

Sunflower stem canker - *Diaporthe/Phomopsis helianthi* (Munt. Cvét. et al.) was firstly recorded in 1980 in sunflower crops in Vojvodina-Yugoslavia (Mihalcevic et al., 1980).

In Romania this pathogen was identified in 1981 in sunflower cropping in the west of the country (Iliescu and Csép, 1982). In 1984, this parasite expanded to the Danube Plain, occurring at present in all sunflower cultivating zones.

The pathogen was described and studied by several authors, starting with Muntanola Cvétkovic, who identified it in 1981 as belonging to *Diaporthe* genus, proposing a binominal name: *Diaporthe/*(perfect form) and *Phomopsis* (imperfect form).

To eliminate damages caused by this parasite, besides phytosanitary, crop and soil management and chemical means, genetic resistance has an outstanding place.

Preliminary results on genetic resistance to the attack by this pathogen have been published by **Vrânceanu** et al., 1983, stating that an important genetic variability exists in response of sunflower genotypes to the attack of this fungus, this leading to an opportunity for improvement of this trait in a genetic way. They showed that the resistance is polygenic (a small number of genes), mostly associated with the "stay green" character of stem (stem remains green at maturity).
Starting from the need to use some forms resistant to the attack by this pathogen in the breeding programme, a great number of sunflower lines and hybrids have been tested in field under natural infection and in field and laboratory under artificial inoculation.

Materials and methods

In 1989-1993 the attack of *Diaporthe/Phomopsis helianthi* (Munt. Cvét. et al.) has been tested at the Research Institute for Cereals and Industrial Crops, Fundulea, on 1250 lines and 310 hybrids with natural infection, and 410 lines and 40 hybrids from these also were tested with artificial inoculation, both in field and laboratory.
Experimental layout was in randomized blocks with 3-5 replications.
Field artificial infection was performed with diseased sunflower stem fragments overwintered on soil, and sprayed with water every 10 days, when winter was droughty. Before sunflower blossoming (flower buds phase) stem fragments were placed on soil at the stem basis, providing with sufficient moisture for contamination by low aspersion rates for 5-7 days.
Other method of artificial infection was inoculation with the fungal mycelium grown on PDA medium, without harming the tissue of host plant (peduncle).
In selection works for resistance to this parasite, a method of artificial infection was used, consisting of germinating sunflower seeds in Petri dishes with filter paper moistened with a solution containing the fungus mycelium.
Scores of infection by the pathogen used a 1-5 scale, adopted in dependence on stem spots, with or without destroying marrow or causing plant lodging (1 - very sensitive, 5 highly resistant).

Results, discussion and conclusion

Reduced rainfall in Fundulea area during June-July, when plants were in flower bud phase and air temperatures especially minima, showed that during the 5 years of studies the best conditions for pathogen setting on plants have been secured in 1991, when rainfall frequency was sufficiently high, while minimal temperatures exceeded 15°C, and maximal ones did not overpass 30°C (Fig.1).
As to air relative humidity in the same period, as related to minimum and maximum temperatures, in 1991 daily values were frequently 80 - 90%, whilst the minimum and maximum temperatures were favourable (Fig.2).

Throughout the 5 experimental years the reaction of sunflower genotypes was pursued and observations effected on infection by this pathogen allowed to include them in 5 resistance classes, as depending on the type of reaction to the attack by the pathogen (Table 1).

Table 1

Reaction of some sunflower genotypes to *Diaporthe (Phomopsis) helianthi* (Munt. Cvét et. al) attack under natural infection conditions Fundulea, 1989-1993

Attack intensity	Resistance class	Reaction type	Lines				Hybrids	
			CMS		RF		F_1	
			Nr.	%	Nr.	%	Nr.	%
1	1	very sensitive	185	23.5	147	31.6	42	13.5
2	2	sensitive	555	70.7	292	62.5	205	66.5
3	3	medium-sensitive	38	4.9	21	4.8	48	15.2
4	4	resistant	7	0.9	5	1.1	15	4.8
5	5	very resistant	0	0	0	0	0	0
Total tested genotypes			785	100	465	100	310	100

Among the sunflower lines studied during 5 years with regard to the attack by *Phomopsis,* the lines AC-1 (LC-1004 A), AC-2 (LC-1020 A), AC-3 and AC-4 (cms lines, used as mother - lines in hybrids) revealed a high resistance degree, and the lines RF-1, RF-2 and RF-3 (fertility restorer lines), these lines being included in the current and prospective hybrids (Table 2).

Study of genetic inheritance of sunflower resistance to the attack by this pathogen proved to be of polygenic nature, being necessary that both mother and father lines be resistant, in order to obtain a resistant hybrid. Their combination in the current and prospective hybrids demonstrated this fact (Table 3).

Notwithstanding, there are cases where a line is resistant and the other medium-resistant, whereas the hybrid is resistant. This outcome depends on the mode of combination of favourable alleles within the hybrid.

According to observations in 1989-1990 a number of sunflower genotypes have been selected for their resistance to *Phomopsis* attack and introduced in a breeding process with a view to obtaining some resistant forms. Selection was performed under heavy artificial infection. After 4 years of selection the resistance level changed, as resulted from Table 4.

In 1992 a comparative set of hybrids was trialled under natural and artificial infection, to seek for the influence of *Phomopsis* attack on seed yield and oil content (Table 5).

The fact is obvious that the hybrids were exposed to high yield losses (seeds and oil) when the resistance level was very low, this difference being more marked in seed oil content.

Table 2

Reaction of some sunflower lines to the attack of *Diaporthe (Phomopsis) helianthi* (Munt. Cvét et al.) under artificial and natural conditions Fundulea, 1990-1993

No.	Inbread line	Attack frequency		Attack intensity		Plant reaction
		N	A	N	A	
1	AC-23	65	57	2	1	S
2	AC-24	41	48	2	2	S
3	RF-10	41	39	2	2	S
4	RF-6	39	49	2	2	S
5	AC-9	32	48	3	3	MR
6	AC-10	29	51	3	3	MR
7	AC-14	31	39	3	3	MR
8	AC-18	23	45	3	3	MR
9	RF-4	29	27	3	3	MR
10	RF-5	30	44	3	3	MR
11	RF-12	34	41	3	3	MR
12	AC-1	12	20	4	4	R
13	AC-2	15	19	4	4	R
14	AC-3	11	27	4	4	R
15	AC-4	17	30	4	4	R
16	RF-1	21	14	4	4	R
17	RF-2	17	18	4	4	R
18	RF-3	25	28	4	4	R
19	Select (Mt. R-1)	17	15	4	4	R
20	F-206 (Mt. S- 2)	65	78	2	1	S

1. Resistant check LSD (P≤ 0.05) = 15.3 (%)
2. Sensitive check

Table 3

Reaction of some sunflower hybrids to *Diaporthe (Phomopsis) helianthi* (Munt. Cvét et al.) attack under natural infection conditions Fundulea, 1990-1993

No.	Hybrid	Attack frequency	Attack intensity	Plant reaction
1	HS-1904 (AC-2A x RF-2)	7	4	R
2	HS-1976 (AC-1A x RF-1)	11	4	R
3	HS-1907 (AC-1A x RF-4)	12	4	R
4	HS-1936 (AC-1A x RF-3)	8	4	R
5	HT-1934 (AC-1A x AC-1B) x RF-3	14	4	R
6	HS-1935 (AC-2A x RF-3)	11	4	R
7	HS-1938 (AC-3A x RF-3)	15	4	R
8	HS-1646 (AC-1A x RF-5)	19	4	R
9	HT-1932 (AC-1A x AC-12B) x RF-3	21	4	R
10	HS-1974 (AC-6A x RF-1)	21	3	MR
11	HS-1952 (AC-5A x RF-3)	24	3	MR
12	HS-1936 (AC-1A x RF-12)	29	3	MR
13	HT-1958 (AC-1A x AC-10B) x RF-12	25	3	MR
14	HS-1881 (AC-1A x RF-18)	31	3	MR
15	SELECT - Check	7	4	R

LSD (P≤ 0.05) = 6.7 (%)

Table 4

Changes of resistance degree after four generations under selection for resistance to *Diaporthe (Phomopsis) helianthi* (Munt. Cvét et al.) in eight sunflower genotypes

No.	Genotype	Attack intensity		Relative plant resistance	
		1990	1993	1990	1993
1	AC - 4	4	5	R	FR
2	RF - 2	4	5	R	FR
3	AC - 11	3	4	MR	R
4	AC - 14	3	4	MR	R
5	AC - 18	3	4	MR	R
6	RF - 8	3	4	MR	R
7	RF - 14	3	4	MR	R
8	RF - 10	3	4	MR	R

Table 5

Seed yield and oil content in sunflower hybrids cropped under conditions of natural and artificial infection with *Diaporthe (Phomopsis) helianthi* (Munt. Cvét et al.) Fundulea, 1992

No.	Hybrid	Attack intensity (I)		Yield (kg/ha)		Oil content (%)	
		nat. inf.	artif. inf.	nat. inf.	artif. inf.	nat. inf.	artif. inf.
1	HS - 1083	4	3	4124	3160	50.1	42.7
2	HT - 1835	5	4	3910	3550	53.0	51.0
3	HT - 1932	5	5	3780	3610	52.3	51.7
4	HS - 1938	5	5	4090	3850	52.8	52.0
5	HT - 1934	5	4	4043	3790	52.1	50.9
6	HS - 1962	4	4	4038	3880	49.5	46.3
7	HS - 1961	4	3	3636	3240	49.0	46.7
8	HS - 1845	3	2	3790	3240	52.9	47.1
9	HS - 1935	4	4	4060	3810	52.7	50.9
10	HS - 1936	4	3	3850	3620	51.8	48.3
11	HS - 1965	2	2	3500	2870	51.2	48.0
12	HS - 1968	4	3	3716	3580	51.9	49.7
13	HS - 1969	4	3	3630	3550	53.1	50.8
14	HS - 2108	4	3	3230	3010	51.2	50.9
15	HS - LV - 206	3	3	3870	3720	48.4	44.9
16	HS - LV - 267	3	3	3840	3550	47.5	42.8
17	PI - 1 / 91	2	1	3520	2620	48.3	43.7
18	SELECT (R)	5	4	4170	4040	54.7	54.0
19	F - 206 (S)	2	1	2570	1970	51.7	46.3

Analysis of correlation between yield losses and level of resistance to the attack by this pathogen, both under natural and artificial infection conditions (Fig. 3 and Fig. 4) showed that the values of correlation coefficients were highly significant, thus confirming the high yield losses at reduced levels of resistance to *Phomopsis* attack.

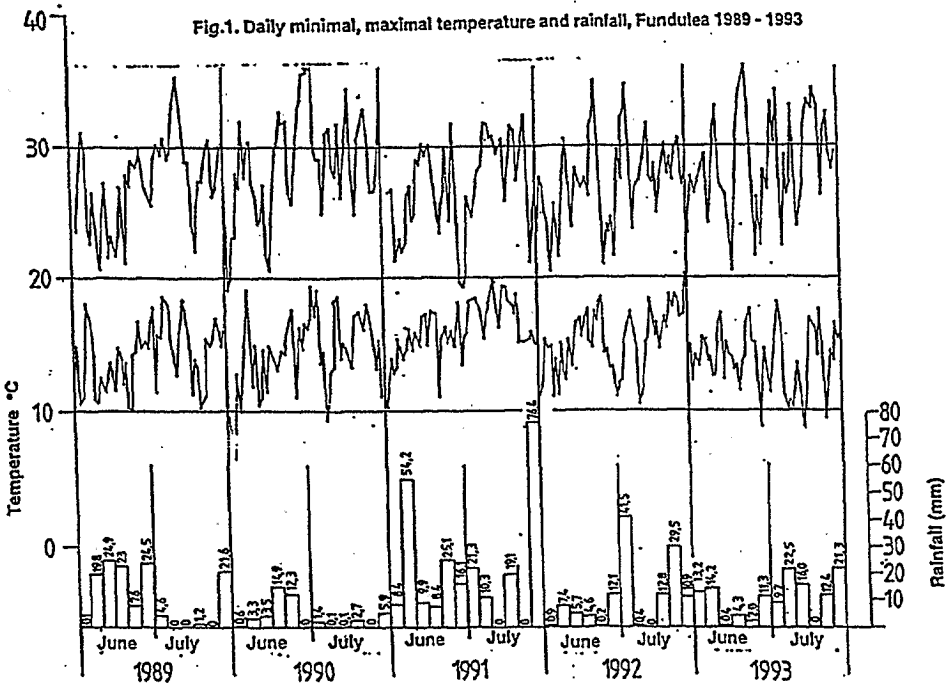

Fig.1. Daily minimal, maximal temperature and rainfall, Fundulea 1989 - 1993

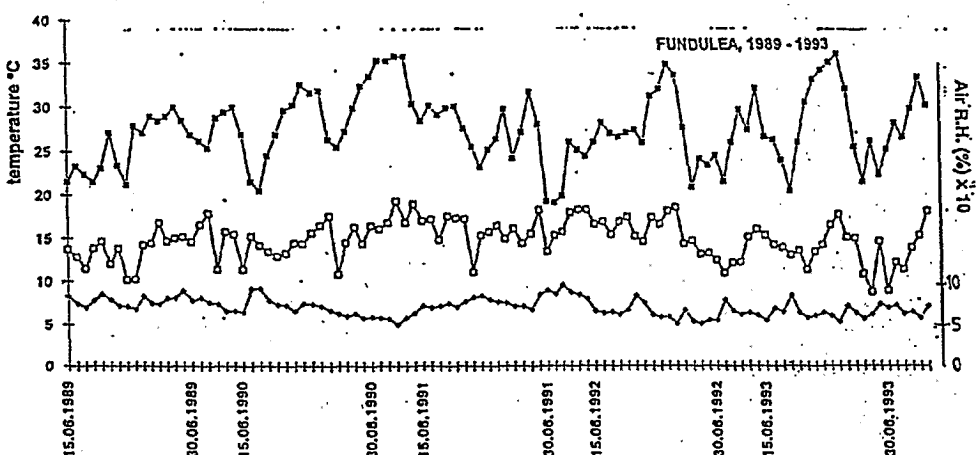

Fig. 2. Daily maximal, minimal temperature and air relative humidity during 15.06. - 05.07

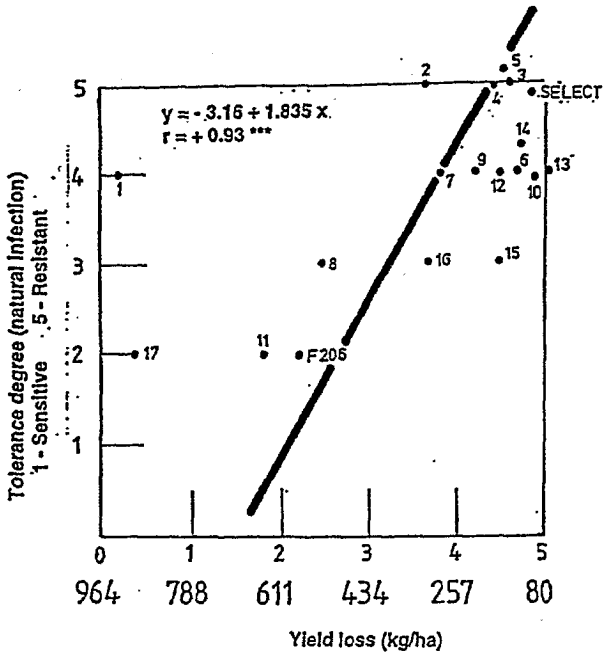

Fig. 3. Correlation between the tolerance degree under natural infection conditions and yield loss

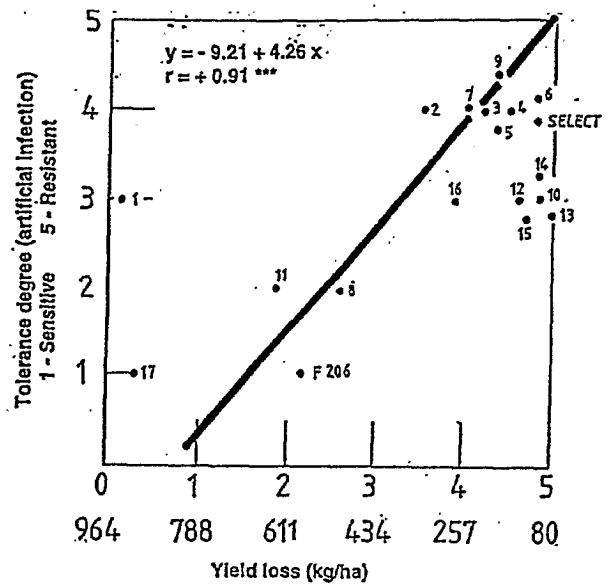

Fig. 4. Correlation between the tolerance degree under artificial infection conditions and yield loss

From the results presented above, the following conclusions evolved:

1. The study of sunflower lines and hybrids under natural and artificial conditions revealed their variable behaviour to the attack of *Diaporthe / Phomopsis helianthi,* a great genetic variability occuring in this respect.
Of the total of trialled lines only 4 cms lines and 3 Rf lines showed high degrees of resistance.

2. Sunflower lines endowed with high resistance degree can be used to create resistant hybrids.

3. Under conditions of heavy artificial infection in population with high resistance degree, highly resistant forms can be obtained by selection.

References

Iliescu H., Csép N., 1982 - Note on the phytosanitary state of sunflower crops in the Western Plain in 1981 (In Romanian), Probl. Prot. Plantelor, Vol. X, p. 91-92.

Mihalcevic M., Muntanola Cvetkovic M., Petrov M., 1980 - Phomopsis sp., a new parasite of sunflower in Yugoslavia. Savremena Poljoprivredna, 28: 531-539.

Muntanola Cvetkovic M., Mihalcevic M., 1981 - On the identity of the causative agent of a serious *Phomopsis / Diaporthe* disease in sunflower plants. Nova Hedwigia, 34: 417-435.

Vrânceanu A.V., Csép N., Pirvu N., Stoenescu F.M., 1983 - Genetic variability of sunflower reaction to the attack of *Phomopsis helianthi* (Munt. Cvét. et. al) Helia, 6: 23-25