STABILITY OF SUNFLOWER RESISTANCE

TO *VERTICILLIUM* WILT

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Summary

We compared the reaction of sunflower hybrids to *Verticillium* wilt (*Verticillium dahliae),* the most important disease of sunflower in Argentina. Our objectives were to detect environmental effects on the reaction of sunflower genotypes to *Verticillium* wilt and to identify non affected-resistant genotypes. We quantified disease intensity in 37 genotypes on the basis of foliage symptoms and stem break in six environments. Four methods were used to assess resistance stability: i) non significant difference with the best genotype in three or more environments, according to a multiple comparison test (BG); ii) reversal interaction between environment and genotype (RI) according to LS Means and graph analysis (the average of the environment was used to test the reversal condition); iii) graphic analysis of the difference between disease intensity means of cultivar and environment. (GA). The disease intensity average of all cultivars tested in one environment represented the environment disease intensity; and iv) standard deviation of the relative disease intensity of a genotype (SDRDI) relative to the environment disease intensity. The environment affected the reaction of the cultivars. Genotypes with good stability and resistance according to BG, RI and GA tests included: CF11, Dekasol 3900, Dekasol 3915, P-64A41, P-64A51, P-64A53, Paraíso 6, Paraíso 20, Pyramid 1 and Zenit. To detect stable and resistance cultivars, it seems promising to start the selection by BG analysis and to run a second selection with the GA method. The use of SDRDI could help to detect levels of stability among stable genotypes.

Introduction

Argentina is the largest exporter of sunflower kernels and oil; 90 % of the harvest it is located in the Pampas Region (Casaburi *et al.*, 1998). The early drying and stem break caused by *Verticillium dahliae* Kleb (*Verticillium* wilt) is the most important disease of sunflower in Argentina (Pereyra & Escande, 1994). The disease causes yield losses up to 73% in highly infested fields (Pereyra *et al*, 1999). Huge losses are due to stem break, however more common are yield losses due to early dying (Pereyra & Escande, 1994).

Microsclerotia of *V. dahliae* survive many years in the soil (Alonso, 1988). The pathogen has a wide host range including 350 plant species, more than 160 families, and important crops like potato, pepper, cotton, olive, sunflower, tomato and alfalfa (Pegg, 1974). The fungus is a natural soil invader that only increases in population by monocropping or rotation with susceptible crops (Alonso, 1988). *Verticillium* wilt management is based on use of cultivar resistance and crop rotations. Every year, our group tests the performance of sunflower cultivars in 12 locations of the South of the province of Buenos Aires and La Pampa, Argentina. Sometimes cultivar response to the disease varies among environments. Our objectives were to confirm if the environment affects the reaction of sunflower genotypes to *V. dahliae* and to identify environmentally stable-resistant genotypes.

Materials and methods

Sunflower genotypes of 37 commercial hybrids were used. Six field crops were established during 1998 and 1999 in the SE of the province of Buenos Aires, Argentina. Soils were naturally infested with *V. dahliae*. In each environment, a complete randomised block design with three replications was used. A plot of one row with 20 plants composed the experimental unit.

At flowering (stage R5 of Schneiter & Miller, 1981), we quantified disease intensity on the basis of foliage symptoms and stem break. A scale of 10 points, based on whole plot assessment, was used. Variance of the data was analysed using a split plot model. Environment was the main plot and cultivar the subplot.

We assessed resistance stability with four methods: i) non significant difference with the best genotype in three or more environments, according to F-protected Least Significant Difference test (BG) (Steel & Torrie, 1993); ii) LS Means analysis of the General Linear Model (SAS Institute, Cary, NC) detected significant differences on *Verticillium* reaction of one cultivar in two environments. The reversal interaction was determined by graph analysis of paired environments where cultivar means were compared with environment means (RI); iii) Graphic analysis of the difference between disease intensity means of cultivar and environment. The disease intensity average of all cultivars tested in one environment represented the environment disease intensity (GA); and iv) Standard deviations of the disease intensity of a cultivar relative to the one of the environment (SDRDI) (Yau & Hamblin, 1994). Standard deviations higher than 40% were arbitrarily considered indication of poor stability.

To test the confidence of the methods, we analysed the significance of the cultivar-environment interaction by variance analysis. In these tests we only included the genotypes characterized as stables by each method.

Results and Discussion

Verticillium wilt intensity in the tested fields oscillated between 3 and 27%. The environment affected the response of the cultivars to the disease. The analysis of variance of the reaction to *V. dahliae* of 37 cultivars in the six environments detected a significant interaction (P=0.0001) between cultivar and environment. The BG analysis selected 14 cultivars (table 1). This group of cultivars did not differ significantly from the most resistant cultivar in at least 3 of 6 environments. The cultivar-environment interaction for these cultivars was significant (P=0.0049), indicating instability of some of the cultivars.

Table 1. Stability of resistance of sunflower cultivars to *Verticillium* wilt determined by four methods.

Cultivar	Best Genotype (BG)	Significant Reversal Interaction (SRI)	Graphic Analysis (GA)	Standard Deviation of Relative Disease Intensity (SDRDI)
Pyramid 1	S	S	S	HS
CF 11	S	S	S	HS
Dekasol	S	S	S	S
Paraíso 20	S	S	S	PS
Paraíso 6	S	S	S	S
P-64A53	S	S	S	S
P-64A51	S	S	S	S
Dekasol	S	S	S	S
Zenit	S	S	S	PS
P-64A41	S	S	S	S
Jagüel	S	S	U	-
GI 720	S	S	U	-
Toba	S	S	U	-
Timbo3	S	U	U	-

S: Stable across environments; U: Unstable across environments; -: Genotypes not included because of GA.
BG: Cultivars without significant difference with the most resistant genotype in at least three of six field environments. Cultivars discarded by this method included ACA 884, Agrobel 960, Agrobel 965, Buck Campoflor, Buck Surcoflor, Cariló, Cauquén, Contiflor 9, Dekasol 3881, DM 226, GI 700M, Kepler, M-742, Rancul, Sankol, SPS 3191, SPS 4530, SPS 7926, TC 3003, TC 3004, Timbo 2, Trident and VDH 480.
RI: LS Means analysis detected significant reaction differences of one cultivar tested in two environments. The reversal interaction was determined by graph analysis of paired environments where cultivar means were compared with environment means.
GA: Graphic analysis of the difference between disease intensity means of cultivar and environment. The disease intensity average of all cultivars tested in one environment represented the environment disease intensity.
SDRDI: Standard deviations of the relative disease intensity of a cultivar (in relation with that of the environment). Arbitrarily SD higher than 40% were considered indication of poor stability. HS: Highly stable; PS: Poorly stable.

The RI method was used to detect reversal cultivar-environment interactions. LS Mean analysis was used to detect differences between the reactions of the same cultivar in paired environments. Because these differences could indicate interactions or just environment effect, we complement the LS Mean test with graphic analysis. RI method found one unstable cultivar (table 1). However, the interaction cultivar-environment of the 13 cultivars detected as stables was almost significant (P=0.0729).

The GA analysis detected four unstable cultivars into the group of 14 stable cultivars according to BG method (Figure 1). Cultivars discarded by this method had large oscillations in disease intensity in relation to the average disease intensity of the environment. The interaction cultivar-environment of the 10 cultivars detected as stables was not significant (P=0.3082). This result supports the suitability of this method to test *Verticillium*-wilt resistance stability of sunflower genotypes.

RI method did not detect three of the interactions found by graph analysis. RI detected large variations of cultivar reaction across environments, while GA detected three cultivars that reduced the probability of a significant interaction from 0.3082 to 0.0729.

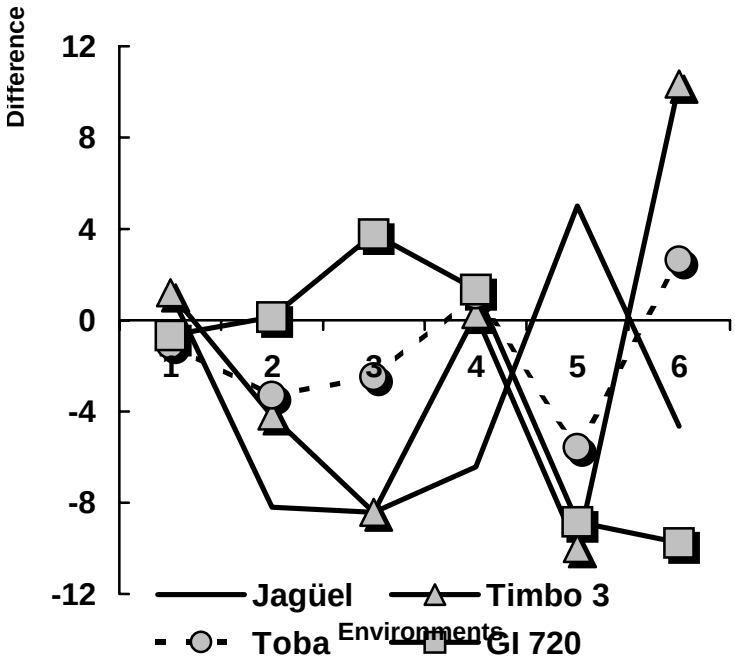

Figure 1. Graphic analysis of the difference between disease intensity of each cultivar and the mean of all cultivars in the corresponding environment. Environments 1 to 6 were Balcarce 98, Belloq 98, Balcarce 99, Pieres 98, Irrigated Balcarce 99 and Pieres 99, respectively. Disease intensity in each environment (average of all 37 cultivars) were 3, 11, 14, 16, 17 and 27, respectively. The horizontal zero line represented the disease intensity average of the environment.

The disease intensity average of the ten cultivars classified as stables was significantly higher in Pieres 99 (19%) than in any other environment (P=0.0001). The significantly lower

disease intensity for the environment (based on these 10 cultivars) was registered in Balcarce 98 (1%).

Finally, to point out the most stable genotypes, we compared the standard deviation of the relative disease intensity of each cultivar across the environments (figure 2). Cultivars with SD higher than 40% were classified as little stable ones.

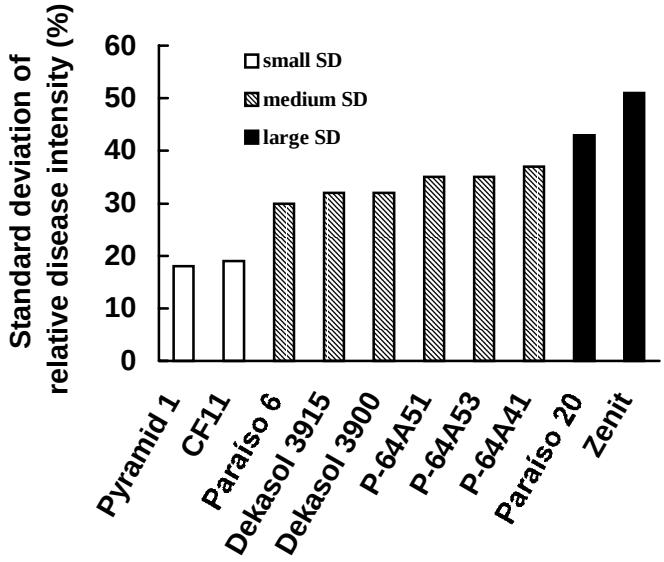

Figure 2. Standard deviation of relative disease intensity of *Verticillium* wilt resistant cultivars. (Relative disease intensity=Cultivar disease intensity/Mean disease intensity of all cultivars expressed as percentage).

Genotypes with good stability and resistance (figures 2 and 3) included: CF11, Dekasol 3900, Dekasol 3915, P-64A41, P-64A51, P-64A53, Paraíso 6, Paraíso 20, Pyramid 1 and Zenit. Cultivars Pyramid 1 and CF 11 were the most stables and resistant.

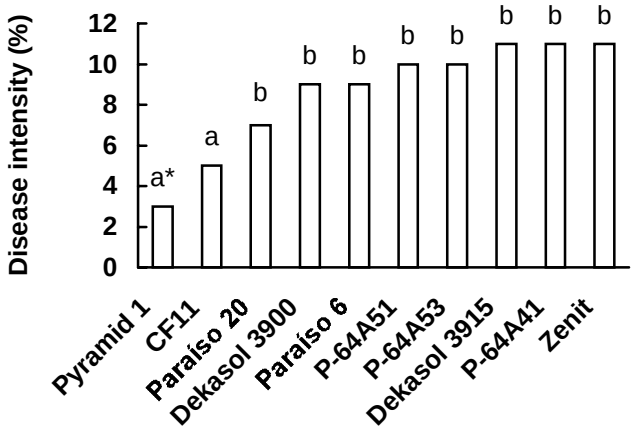

Figure 3. Disease intensity of *Verticillium* dahliae in stable resistant cultivars.
* Columns with the same letter did not differ significantly (P=0.0001).

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