

VARIABILITY OF *Sclerotinia* RESPONSES IN *Helianthus petiolaris*

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SUMMARY

Cultivated sunflower (*Helianthus annuus* L.) is susceptible to *Sclerotinia sclerotiorum* (Lib.) de Bary. *H. petiolaris* Nutt is a wild sunflower species naturalized in Argentina. The objective of this work was to evaluate the responses of some accessions of *H. petiolaris* to *S. sclerotiorum* inoculation in order to detect potential sources of resistance to be used in cultivated sunflower. An experiment was carried out at the UIB's experiment field. Inoculations were made on leaves and stems. Lesion mean on leaves was 22 mm and 24 mm on stems. The accession GRR-252 showed mean lesions on leaves significantly longer than in GRR-250. However, there were no differences for stem inoculation. Correlation analysis indicated the independence of leaf and stem responses. Results obtained after the first experimental trial suggest the possibility of detecting some *H. petiolaris* accessions with a higher level of resistance to *S. sclerotiorum* than others.

Key words: *Helianthus petiolaris*, leaf, resistance test, *Sclerotinia sclerotiorum*, stem, wild sunflower

INTRODUCTION

Sunflower is one of the most important annual crops cultivated for its edible oil. *Sclerotinia sclerotiorum* is a severe limitation because seed yield and seed oil content of this oleaginous crop can be drastically reduced by the pathogen (Gulya *et al.*, 1997).

The genus *Helianthus* contains 49 species and most of them have been evaluated in order to detect potential sources of resistance which might be utilized in cultivated sunflower breeding programs (Fick and Miller, 1997).

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In Argentina some wild species of sunflower are naturalized, particularly *Helianthus petiolaris*. This wild species is distributed in the large cropping area of Argentina which includes five provinces, Buenos Aires, La Pampa, Córdoba, San Luis and San Juan (Covas *et al.*, 1970; Poverene *et al.*, 2004).

Certain wild species of sunflower or their interspecific progenies after crossing with *H. annuus* have shown high levels of resistance to *S. sclerotiorum* (Christov *et al.*, 1996; Cerboncini *et al.*, 2002, 2005; Rashid and Seiler, 2006). Christov *et al.* (1996) also indicated the wild *H. petiolaris* as having resistance genes to downy mildew caused by *Plasmopara halstedii* infection.

No work has been found evaluating *H. petiolaris* responses to *Sclerotinia* resistance. Therefore, it seemed worthwhile to evaluate this large array of genetic resources in Argentina in order to identify genotypes with good performance, for subsequent transfer of that resistance to cultivated sunflower.

The objective of this work was to assess a series of *H. petiolaris* accessions, naturalized in Argentina, in order to detect potential sources of resistance to *S. sclerotiorum* infection.

MATERIALS AND METHODS

Inflorescences of 105 plants of *H. petiolaris* were collected from 5 different sites situated between the coordinates 36°32'33"-37°08'15" S, and 64°00'55"-63°02'55" W, in the Provinces of Buenos Aires and La Pampa, Argentina. Plants from a unique collection site were considered as an *H. petiolaris* accession, which were introduced and then classified according to a number given by the Germplasm Bank at the EEA Balcarce-INTA. All capitula of each plant were threshed and seeds bulked and stored during four months at 5°C. Seeds from each plant represented a half-sib family (HSF) and there were different numbers of them, varying from 10 to 33 per accession (Tables 1 and 2).

Seeds were grown under greenhouse conditions and two HSF were then rejected due to germination problems. Seedlings of 20 days were transplanted at the UIB's experiment field following a randomized complete block design with two replicates.

Inoculations on leaves and stems were made 60 and 68 days after transplanting, respectively, using an *S. sclerotiorum* isolate of well-known aggressiveness (Calaza *et al.*, 2005). Inoculations were made following a protocol used in cultivated sunflower (Castaño *et al.*, 1993). Agar disks, 5 mm diameter and containing the pathogen mycelium, were placed on two leaves/plant in 50% of plants selected in the plot. Two stems/plant were inoculated in the remaining plants. Lesion lengths were estimated on the central nervure of leaves and above and below the infection site on stems, 6 and 4 days after inoculation, respectively.

Only lesion data greater than 5 mm were used for statistical analysis. In the analysis of variance a two-factor mixed model was considered, where the source of variation "accessions" were considered as fixed effect, while "HSF/accession" were random effect. All analyses were based on Steel and Torrie (1988).

RESULTS AND DISCUSSION

Lesions of *S. sclerotiorum* in *H. petiolaris* were similar to those produced by the same pathogen after inoculation of the cultivated sunflower (Figures 1 and 2).



Figure 1: Light brown lesion produced by *Sclerotinia* inoculation of *H. petiolaris* leaf

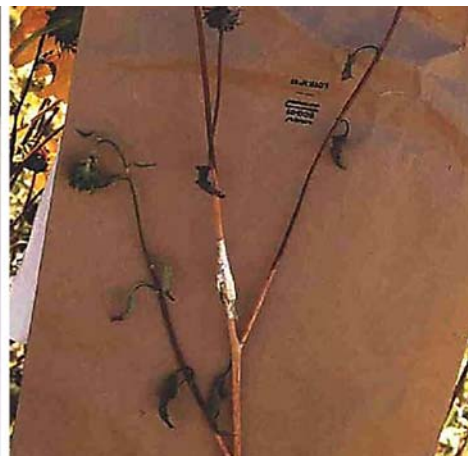


Figure 2: Light brown lesion produced by *Sclerotinia* inoculation of *H. petiolaris* mid-stem

Lesion lengths on leaves greater than 5 mm were observed in 93.7% (1002/1069) of inoculated plants and in 94.2% (1658/1759) of leaves showing disease symptoms (Table 1). Mean lesion length was 22 mm and symptoms ranged from 12.5 to 33 mm in 103 HSFs. The coefficient of variability, CV=44%, suggested a relatively low precision of the data.

Table 1: Descriptive statistical measures in 5 accessions of *H. petiolaris* after *S. sclerotiorum* inoculation of leaves

Accessions	No. of inoculated plants	No. of leaves with symptoms	No. of leaves with symptoms > 5 mm	No. of plants with symptoms > 5 mm	No. of half-sib families	Lesions on leaves	
						Means (mm)	Range (mm) Min. Max.
GRR-250	378	592	571	348	33	20.2	13.5 29.3
GRR-252	167	280	264	159	19	24.7	18.6 33
GRR-254	218	341	305	197	17	22.4	16.6 29.8
GRR-255	220	390	366	212	24	22	12.5 30
GRR-256	86	156	152	86	10	20.9	18.1 27.3
Totals	1069	1759	1658	1002	103	22	

LSD_{0.05}=2.2

Analysis of variance detected large differences ($p < 0.001$) among the *H. petiolaris* accessions. Given a LSD value of 2.2 mm, the accession GRR-250, with the minimum mean value of lesions (20.2 mm), showed a significantly ($p < 0.001$) higher level of resistance than GRR-252 (24.7 mm). However, there were no detected differences in HSF/accessions.

Symptoms larger than 5 mm were shown by 93.2% (919/986) of the plants with inoculated stems, while 91.7% (1576/1719) of stems registered lesion lengths larger to 5 mm (Table 2).

Table 2: Descriptive statistical measures in 5 accessions of *H. petiolaris* after *S. sclerotiorum* inoculation of stems.

Accessions	No. of inoculated plants	No. of stems with symptoms	No. of stems with symptoms > 5 mm	No. of plants with symptoms > 5 mm	No. of half-sib families	Lesions on stems	
						Means (mm)	Range (mm) Min. Max.
GRR-250	353	596	548	325	33	23.6	8.5 49.9
GRR-252	153	259	234	140	19	19.7	8 49.5
GRR-254	197	360	334	191	17	24.2	14 44.3
GRR-255	199	350	318	181	24	27.4	7 82
GRR-256	84	154	142	82	10	26.1	13.2 36.6
Totals	986	1719	1576	919	103	24.2	

LSD_{0.05}=2.4

The average of stem lesions was 24.2 mm and symptoms ranged from 7 to 82 mm. Analysis of variance showed that *H. petiolaris* accessions did not differ in mean responses after the inoculation of stems. Despite the relatively low precision of the analysis (CV=75%), it was nevertheless possible to detect significant ($p < 0.001$) differences in HSF/accession.

LSD test was made and results are shown in Table 3. LSD values allowed detection of HSF/accession having mean lesion lengths on the stem similar to those with the minimum value. In this sense, the proportion of HSFs with best performances by accession varied between 50 and 84%.

Table 3: Least significant differences in HSF/accessions of *H. petiolaris* stems inoculated with *S. sclerotiorum*.

Accessions	Lowest mean lesion on stems (mm)	LSD _{0.05} value (mm)	Families with best performance	
			No	%
GRR-250	8.5	18.3	22	67
GRR-252	8.5	25.9	16	84
GRR-254	13.9	16.3	13	76
GRR-255	7	29.5	19	79
GRR-256	13.2	17.3	5	50

The Spearman's correlation coefficient of $r_s = 0.16$, no different from zero, indicated that the responses of *H. petiolaris* inoculated on leaves and stems were independent. This result agrees with Castaño *et al.* (1993) who showed a lack of

association between the responses to inoculation made in the same organs, but on cultivated sunflowers.

Further studies are needed to evaluate the repeatability of *H. petiolaris* responses to *S. sclerotiorum* inoculations of leaves and stems. In spite of this, it should be mentioned that some genotypes had better levels of resistance than others after *S. sclerotiorum* inoculation of leaves and stems. This germplasm could be considered as a potential source of resistance in breeding sunflower plans for disease resistance.

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VARIABILIDAD DE LAS RESPUESTAS DE *Helianthus petiolaris* A LAS INFECCIONES DE *Sclerotinia sclerotiorum*

RESUMEN

El girasol cultivado (*Helianthus annuus*) es susceptible a las infecciones de *Sclerotinia sclerotiorum* y la especie silvestre *H. petiolaris* está naturalizada en la Argentina. El objetivo de este trabajo fue evaluar algunas introducciones de *H. petiolaris*, mediante inoculaciones de *S. sclerotiorum*, a fin de detectar potenciales fuentes de resistencia que puedan ser utilizables en el girasol cultivado. Se realizó un experimento en el campo experimental de la UIB. Las inoculaciones se realizaron en hojas y tallos de plantas elegidas adrede. Se obtuvo un promedio de lesión por parcela. La lesión media en hojas fue de 22 mm, mientras que en tallos fue 24 mm. En las hojas, fue posible diferenciar a las introducciones por su comportamiento medio. El valor del test LSD determinó que la lesión promedio de la introducción GRR-252 fue significativamente mayor que la de GRR-250. Para la infección en tallos no hubo diferencias. El coeficiente de correlación de rangos entre las lesiones producidas en cada órgano no fue significativo. Este primer año de resultados indicaría la posibilidad de detectar introducciones con comportamientos diferenciales a *S. sclerotiorum*.

VARIABILITÉ DES RÉPONSES D'*Helianthus petiolaris* FACE AUX INFECTIONS DE *Sclerotinia sclerotiorum*

RÉSUMÉ

Le tournesol cultivé (*Helianthus annuus*) est sensible au *Sclerotinia sclerotiorum*. L' *H. petiolaris* est une des espèces sauvages de tournesol qui est naturalisée en Argentine. L'objectif de ce travail est d'évaluer quelques introductions d'*H. petiolaris*, face aux infections de *S. sclerotiorum*, afin de détecter des sources de résistance potentielle pour le tournesol cultivé. Une expérience a été mise en place dans l'UIB. Les infections ont été faites en feuilles ainsi qu'en tiges des plantes de *H. petiolaris*. La moyenne des réponses en feuilles était 22 millimètres, tandis qu'aux tiges était 24 millimètres. En feuilles, des différences significatives ont été détectées parmi les introductions et la valeur de la plus petite différence significative a déterminé que l'introduction GRR-252 avait un niveau de résistance significativement plus bas que GRR-250. En tiges il n'y a cependant aucune différence parmi les génotypes. Le coefficient de corrélation des rangs parmi les réponses feuilles-tiges n'était non plus significatif. Cette première année des résultats indiquerait la possibilité de détecter des génotypes d'*H. petiolaris* de bon comportement face au *S. sclerotiorum*.

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