Inheritance of partial resistance to black stem (*Phoma macdonaldii*) in sunflower

A. Roustaee[a], G. Barrault[a], G. Dechamp-Guillaume[a], P. Lesigne[b] and A. Sarrafi[a]*†

[a]*Laboratoire de Biotechnologie et Amélioration des Plantes, PBV, INP-ENSAT, UA INRA, 18 chemin de Borde Rouge, BP 107, F-31326 Castanet Cedex, France; and* [b]*ASGROW SA, Centre de Recherche et de Développement, Zone Maraichère 'Les Sablons', BP 5, 37270 Saint Martin Le Beau, France*

Two programmes to investigate the inheritance of resistance to black stem in sunflowers were undertaken in a controlled-environment growth chamber. In the first, an experiment was performed using a randomized complete block design with 24 lines, six male-sterile (A lines), six maintainers (B lines), six restorers (R lines) and their six F_1 hybrids in six replications. Each treatment consisted of 12 seedlings. Twelve-day-old seedlings were inoculated with a suspension of pycniospores, and 7 days later the two cotyledon petioles of each seedling were scored on a 1–9 scale for the percentage of necrotic area. Some alloplasmic lines (which have the same nucleus and different cytoplasms) gave a significant cytoplasmic effect for improved partial resistance to the disease. Deviation of F_1 hybrids from the mean of the parent values was significant for partial resistance to *Phoma macdonaldii* in three of the six F_1 hybrids. Two further experiments with eight lines (resistant and susceptible) at the fifth leaf-pair and flowering stages were carried out under the same conditions. At both growth stages the previous classification of lines at the seedling stage was confirmed. In the second programme, five male-sterile sunflower lines were crossed with five fertility-restorers in a factorial mating design. The 10 inbred lines and their 25 F_1 hybrids were studied in two successive experiments under the same conditions and with the same experimental design and isolate of *Phoma* as in the seedling-stage experiment in the first programme. Analysis of variance showed that male-sterile and restorer lines possessed general combining abilities, and also that specific combining abilities of F_1 hybrids were significant. The estimates of general combining ability for partial resistance were significant in AS617A, AS618A and AS614R inbred lines. These lines are available for developing F_1 hybrids with improved resistance to *Phoma* in sunflower-breeding programmes.

Keywords: black stem, combining ability, partial resistance, *Phoma macdonaldii*, sunflower

Introduction

Black stem caused by *Leptosphaeria lindquistiii* (*Phoma macdonaldii*) is one of the most important diseases of sunflower. It is present in many European countries, including Yugoslavia, Italy, Romania and Bulgaria (Acimovic, 1984), China (Hua & Ma, 1996), Australia and the USA (Acimovic, 1984). Since 1990 the disease has continued to spread and is now recognized as one of the most serious diseases of sunflower in France (Peres & Lefol, 1996).

The symptoms appear principally on the stems and petioles. Disease lesions usually begin at the base of the petiole and spread rapidly to form a large black area on the stem. Premature ripening of diseased plants causes a 10–30% reduction in yield (Penaud, 1996) and 1000-seed weight (Carson, 1991). It can also cause premature death of the plant (Donald *et al*., 1987).

Estimates of genetic variation and combining ability are useful in determining the breeding value of lines and the appropriate procedures to use in a breeding programme. General combining ability (GCA) is an important indicator of the value of inbred lines in hybrid combinations. Differences in GCA are attributed mainly to the effects of additive genes, whereas differences in specific combining abilities (SCA) have been attributed to nonadditive genetic variance (Falconer, 1972).

Genetic studies of partial resistance to a disease require a standardized disease-scoring system that must be rapid and purely visual. Scoring the damage caused by the pathogen in naturally infected lines under field conditions can be reliable, but it is not always possible to expose plants to the pathogen uniformly and so achieve uniform infection. For this reason, different methods of inoculation with a pycniospore suspension

*To whom correspondence should be addressed.

†E-mail: sarrafi@ensat.fr

on leaf petioles in oilseed rape (Brunin & Lacoste, 1970; Newman & Bailey, 1987) and other crucifers (Williams & Delwiche, 1979) have been proposed, which are valuable for screening large numbers of lines. In the present work a method was developed using artificially controlled conditions for inoculation of sunflower seedlings with *Phoma*. This method can be used for screening lines and for genetic studies of black stem in sunflower.

Genetic variability for partial resistance to black stem in sunflower has been observed under field conditions (Peres *et al*., 1994). The authors are not aware of any previous reports of the inheritance of partial resistance to black stem in sunflower. The aim of this study was to determine the genetic control of partial resistance to *Phoma* in sunflower using 28 inbred lines showing a range of susceptibility and their 31 F_1 hybrids, in two different programmes.

Materials and methods

First programme

An experiment was carried out with 24 lines of sunflower, including six cytoplasmic male-sterile inbred lines used as female (ASA1, ASA2, ASA3, ASA4, ASA5 and ASA6), the same six inbred lines with their own normal cytoplasm (maintainer lines), which are male-fertile (ASB1, ASB2, ASB3, ASB4, ASB5 and ASB6), six restorer inbred lines used as male (ASR1, ASR2, ASR3, ASR4, ASR5 and ASR6) and six F_1 hybrids from crosses between male-sterile and restorer lines (A1 × R1, A2 × R2, A3 × R3, A4 × R4, A5 × R5 and A6 × R6). These F_1 hybrids and their parents represent a high level of genetic diversity and were selected by ASGROW France SA on the basis of their productivity.

A monopycniospore isolate of *Phoma*, produced by this department from naturally infected plants in south-west France, was used in the study. The pathological and physiological characteristics of this isolate, which is one of the most aggressive isolates of the pathogen, were kept constant during the period of the experiment using the method of conservation described by Arabi *et al*. (1992) for *Drechslera teres*.

The experiment was conducted in a controlled-environment chamber at 25 ± 1°C (day)/18 ± 1°C (night) and a relative humidity of 75–80%. Light intensity was 200 μE m^{-2} s^{-1} with a 14 h photoperiod. The experiment was designed as a randomized complete block with six replicates. Each replicate consisted of 12 seedlings of each line. Seeds were sterilized for 5 min in a 6% sodium hypochlorite solution and washed in sterile distilled water. Two rows of six seeds per line per replication were sown in plastic containers (40 × 30 × 30 cm) filled with moistened vermiculite. Plantlets were irrigated with a nutrient solution (NPK 6 : 3 : 6 and micronutrients; Substral, Boulogne Billancourt, France).

Twelve-day-old seedlings were inoculated at the junction of the cotyledon petiole and hypocotyl with 20 μL of a pycniospore suspension (10^6 pycniospores (py) per mL water containing 0·25% gelatine) using a micropipette. After inoculation, each container was enclosed for 72 h using a special transparent cover (plexiglass) to maintain a near-saturated humidity favourable for fungal infection.

In order to compare the partial resistance of lines at the seedling stage with the reaction of adult plants, the most resistant group (ASA4, ASR4, F_1 and ASB4) and the most susceptible group (ASA6, ASR6, F_1 and ASB6) of lines in the seedling experiment were chosen. Their susceptibility to *P. macdonaldii* at the fifth leaf-pair stage and flowering stage, under the same conditions as those used for the seedling stage, was studied in two experiments. At both growth stages, the four youngest leaves of the plant were inoculated at the intersection of petiole and stem with 40 μL of 10^6 py mL^{-1} of isolate MP6, and scored 14 days later.

In the seedling experiment, small chlorotic lesions appeared on the surface of the cotyledon petiole 1–2 days after inoculation. Three days later they had elongated and transformed into necrotic lesions, depending on the reaction of the lines. In severe infection of susceptible lines, the necrotic lesions elongated and then spread down the hypocotyl. Thus the percentage of surface area in the upper part of the cotyledon petiole occupied by the fungus varied with the susceptibility of the line. Seven days after inoculation both cotyledon petioles of the seedling were scored according to the percentage of the petiole area exhibiting disease symptoms (necrosis). A rating scale from 1 to 9, based on the percentage of infected cotyledon petiole area, was used, where 1 = 0–5%, 2 = 6–10%, 3 = 11–20%, 4 = 21–30%, 5 = 31–40%, 6 = 41–60%, 7 = 61–80%, 8 = 81–99% and 9 = 100% with necrosis spreading down the stem. In the other two experiments (fifth leaf-pair and flowering stages) 14 days after inoculation the rating scale used was from 1 to 9, based on the percentage area of infection on the leaf petiole and the form and size of necrosis on the stem, where 1 = 0–10%, 2 = 11–20%, 3 = 21–30%, 4 = 31–40% (scores from 1 to 4 indicate the area of infection on the leaf petiole), 5 = 41–50% (infected petiole area and beginning of necrotic lesions on the stem), 6 = diameter of necrosis on the stem more than 1 cm, 7 = necrosis surrounding the stem, 8 = size of necrosis on the stem more than 2 cm, and 9 = necrosis developed between two successive petioles on the stem. Intensity measures in all experiments needed no transformation to normalize the distribution. Analysis of variance of the complete randomized block was performed. The Newman–Keuls test was used for comparing the mean performances of lines.

Second programme

Two successive experiments were carried out with 35 lines: five male-sterile inbred lines used as females (AS616A, AS617A, AS618A, AS619A and AS621A),

five restorer inbred lines used as males (AS610R, AS612R, AS613R, AS614R and AS615R) and 25 F_1 hybrids from crosses between male-sterile and restorer lines. These F_1 hybrids and their parents represent a high level of genetic diversity and were selected by ASGROW France SA on the basis of their agronomic traits. Experiments were conducted under the same conditions and with the same experimental design and isolate of *Phoma* as in the first programme. Measures of severity were taken at the seedling stage as in the first programme. Mean scores for resistance of the 10 inbred lines were compared using the Newman–Keuls test after ANOVA. Sums of squares for the F_1 hybrids in a factorial analysis were partitioned into male-sterile and restorer effects and male-sterile–restorer interaction effect. A fixed model was assumed and the mean squares for male-sterile, restorer and interaction were tested by the error mean square. The combined main effect of male-sterile and restorer was considered as the GCA effect, and the male-sterile–restorer interaction was equivalent to the SCA effect.

Heritability of resistance to *Phoma*, which estimates the predictability between inbred lines and F_1 hybrids, was assessed by the linear regression coefficient (b) between the mean value of the parents (x) and the value of the corresponding F_1 hybrid (y) (Falconer, 1972).

Results

First programme

ANOVA showed that the effect of line was significant for partial resistance to *P. macdonaldii* at the seedling stage, at the fifth leaf-pair stage and the flowering stage (data not presented). Mean levels of partial resistance to *Phoma* in 18 inbred lines and their six F_1 hybrids are presented in Table 1 and Table 2. The most resistant line was the B line ASB4 (score 2·41) and the most susceptible was ASB3 (7·74) in all experiments (seedling, fifth leaf-pair and flowering stages). Some other lines, such as ASR2, ASA1 and ASB6, displayed medium partial resistance with scores of 4–5.

In six alloplasmic lines (ASA1 and ASB1, ASA4 and ASB4, ASA6 and ASB6, each pair with the same nucleus and different cytoplasms), the male B line (maintainer) with *Helianthus annuus* cytoplasm was significantly more resistant to *Phoma* than the female A line (male-sterile) with *Helianthus petiolaris* cytoplasm (Tables 1 and 2). In contrast, the A line was more resistant than the B line for the pair ASA3 and ASB3, and there was no significant difference between ASA2 and ASB2 (Table 1). These results suggest possible residual effects of B lines (maintainer) and nuclear–cytoplasmic interaction. The A lines were obtained after seven back-crosses.

The genetic variability of F_1 hybrids for partial resistance to *Phoma* is presented in Tables 1 and 2. It is clear that there was a continuous range of susceptibility among the F_1 hybrids from resistant (e.g. ASA2 × ASR2-F_1 with a score of about 3) to very susceptible (e.g. ASA6 × ASR6-F_1 with a score of more than 6). Deviation of F_1 hybrids from the mean of the parents was significant in three F_1 hybrids at the seedling stage, i.e. ASA2 × ASR2-F_1, ASA3 × ASR3-F_1 and ASA4 × ASR4-F_1 (Table 1), with a disease reduction of between 20 and 40%. In the three other F_1 hybrids, and also at the fifth leaf-pair and flowering stages, the percentage deviation was not significant (Tables 1 and 2). The absence of a significant difference between the F_1 hybrid and the average value of their parents for partial resistance to black stem shows the importance of additive genetic control in the material. However, for F_1 hybrids with a significant decrease in susceptibility, resistance appears to be incompletely dominant.

Correlation coefficients between partial resistance values at the fifth leaf-pair and flowering stages with the seedling stage values were positive and significant ($r = 0{\cdot}963$ and 0·927, respectively). Line classification was similar in all experiments. In addition, six commercial F_1 hybrids were tested in the field and growth chamber. Preliminary results showed a good correlation between seedling stage and adult plant susceptibility.

Table 1 Partial resistance to black stem in male-sterile (A line), maintainer (B line), restorer (R line) and F_1 hybrids of sunflower inoculated at the seedling stage

Line	Host reaction	F_1	MP[a]	D[b] (%)
ASA1	5·14 def	5·80 bcd	5·20	11·53 NS
ASB1	3·12 ij			
ASA1–ASB1[c]	2·02*			
ASR1	5·26 cde			
ASA2	4·85 efg	3·07 ij	5·21	−41·07*
ASB2	4·07 hi			
ASA2–ASB2	0·78 NS			
ASR2	5·57 bcd			
ASA3	6·42 abc	5·31 cde	6·66	−20·27*
ASB3	7·74 a			
ASA3–ASB3	−1·32*			
ASR3	6·91 ab			
ASA4	4·49 fgh	4·18 ghi	5·34	−21·72*
ASB4	2·41 j			
ASA4–ASB4	2·08*			
ASR4	6·20 bcd			
ASA5	6·61 abc	4·72 fgh	5·49	−14·02 NS
ASB5	6·41 abc			
ASA5–ASB5	0·2 NS			
ASR5	4·38 fgh			
ASA6	6·41 abc	6·14 bcd	6·54	−6·11 NS
ASB6	4·83 efg			
ASA6–ASB6	1·58*			
ASR6	6·68 abc			

Means in the same column followed by different letters are significantly different at $P = 0{\cdot}05$ (Newman–Keuls test). The values represent the mean of host reaction, scale range from 1 to 9, in six replications, each containing 24 cotyledon petioles.

*Significant at $P = 0{\cdot}05$.

NS, Not significant.

[a]MP, mean of parents = (female A + male R) / 2.

[b]D, percentage deviation of F_1 from mean of parents = (F_1 − MP) x 100 / MP.

Table 2 Partial resistance to black stem in male-sterile (A line), maintainer (B line), restorer (R line) and F_1 hybrids of sunflower inoculated at the fifth leaf-pair and flowering stages

Line	5th LPS[a]	Flowering
ASA4	5·00 b	6·00 abc
ASB4	3·00 c	4·00 c
ASA4-ASB4[b]	2·00*	2·00*
ASR4	5·67 ab	6·30 ab
F_1	4·63 b	4·67 bc
MP[c]	5·33	6·15
D[d]%	−13·12 NS	−24·06 NS
ASA6	6·67 a	7·67 a
ASB6	4·67 b	6·00 abc
ASA6-ASB6	2·00*	1·67*
ASR6	6·33 ab	7·00 ab
F_1	5·67 ab	6·67 ab
MP	6·50	7·33
D%	−12·76 NS	−9·06 NS

Means in the same column followed by different letters are significantly different at $P = 0{\cdot}05$ (Newman–Keuls test). The values represent the mean of host reaction, scale range from 1 to 9, in three replications, each containing 24 leaf petioles and stems.

*Significant at $P = 0{\cdot}05$.

NS, not significant.

[a]5th LPS, fifth leaf-pair stage.

[b]Difference between the two lines.

[c]MP, mean of parents = (female A + male R) / 2.

[d]*D*, percentage deviation of F_1 hybrid from mean of parents = (F_1 − MP) x 100 / MP.

Second programme

ANOVA for the 10 inbred lines showed that the effect of line was significant for partial resistance to black stem at the seedling stage in both experiments (analysis not presented). The most resistant line was the restorer line AS614R (scores of 3·27 and 2·64 in the two experiments) (Table 3), and the most susceptible was the male-sterile line AS616A (scores of 7·43 and 7·24).

Table 3 Mean scores and general combining ability (GCA) of 10 parental inbred lines of sunflower for partial resistance to *Phoma*

	First experiment		Second experiment	
Lines	Mean[a]	GCA	Mean	GCA
Male steriles				
AS616A	7·43 a	0·782*	7·24 a	0·868*
AS617A	5·60 ab	− 0·060	4·26 c	− 0·266
AS618A	5·48 ab	− 0·546*	5·27 b	− 0·418*
AS619A	4·07 bc	− 0·516*	3·94 c	− 0·570*
AS621A	7·19 a	0·338*	6·60 ab	0·384*
Restorers				
AS610R	6·29 a	0·316*	6·24 ab	0·570*
AS612R	6·92 a	0·166	7·12 a	0·032
AS613R	5·88 ab	0·284*	5·94 ab	0·268*
AS614R	3·27 c	− 0·748*	2·64 d	− 0·846*
AS615R	5·83 ab	− 0·020	5·60 b	− 0·026

[a]Means in the same column followed by different letters are significantly different at $P = 0{\cdot}05$ (Newman–Keuls test). Values represent the mean of host reaction, scale range from 1 to 9, from three replications, each containing 24 petioles.

*Significant at $P = 0{\cdot}05$.

Table 4 Mean squares (MS) for a factorial ANOVA for partial resistance to *Phoma* in 25 F_1 hybrids of sunflower

Source of variation	d.f.	First experiment (MS)	Second experiment (MS)
Total	74	1·17	1·55
Male-sterile (GCA)	4	4·86***	6·12***
Restorer (GCA)	4	2·88***	4·04***
M.sterile × Restorer (SCA)	16	2·39***	3·97***
Block	2	0·43	0·22
Residual	48	0·34	0·21

d.f., Degrees of freedom.

***Significant at $P = 0{\cdot}001$.

The GCA of the inbred lines and the SCA of the F_1 hybrids were significant (Table 4). The estimates of GCA effects, which are important indicators of the value of inbred lines in hybrid combinations, were significant and negative (more resistance) for AS618A, AS619A and AS614R in both experiments (Table 3). Estimates of GCA were significant and positive (more susceptibility) for AS616A, AS621A, AS610R and AS613R. Some other inbred lines gave nonsignificant GCA values and the level of resistance of their hybrids was neither increased nor decreased.

SCA values (Table 5) were significant and negative for some of the F_1 hybrids, such as AS616A × AS610R and AS619A × AS612R. These F_1 hybrids have a positive gene interaction in the control of resistance to black stem. Some F_1 hybrids gave significant positive SCA values, indicating that the gene interaction for resistance was negative, for example AS617A × AS613R and AS619A × AS614R.

The heritability estimated from the regression coefficient between mid-parent (x) and F_1 hybrid (y) values was 0·664, which indicates that the resistance level varies in a similar manner in inbreds and hybrids (Fig. 1). However, only 33·4% ($R^2 = 0{\cdot}334$, significant at $P = 0{\cdot}01$) of the partial resistance of F_1 hybrids can be predicted from the resistance level of parents.

The significant interaction between the restorers and male-sterile lines in the ANOVA (Table 4) probably arises from the deviation of F_1 hybrids from the mean of their parents (Fig. 1).

Discussion

This work supports earlier reports of resistance and susceptibility to *Phoma* in sunflower (Peres *et al*., 1994), and provides evidence for the inheritance of resistance in F_1 hybrids and estimates of the combining ability of lines. In some alloplasmic lines, the male line with *H. annuus* cytoplasm was significantly more resistant to *Phoma* than the female line with *H. petiolaris* cytoplasm, suggesting a nuclear–cytoplasmic interaction (Table 1).

The significant negative (more resistant) GCAs for

Table 5 Mean scores and specific combining ability (SCA) of 25 F_1 hybrids for resistance to *Phoma* in a factorial mating design

Restorers										
AS610R		AS612R		AS613R		AS614R		AS615R		
Male steriles	Mean[a]	SCA	Mean	SCA	Mean	SCA	Mean	SCA	Mean	SCA
First experiment										
AS616A	5·20 cde	−1·352*	7·15 a	0·748*	6·43 abc	−0·090	5·27 cde	−0·218	7·13 a	0·914*
AS617A	6·04 abc	0·330	5·79 abc	0·230	6·29 abc	0·612*	3·89 fg	−0·756*	4·96 cde	−0·414
AS618A	5·38 bcd	0·156	5·16 cde	0·086	4·67 def	−0·522	4·50 ef	0·340	4·83 cde	−0·058
AS619A	6·11 abc	0·856*	3·05 g	−2·054*	5·33 bcd	0·108	5·36 bcd	1·170*	4·84 cde	−0·078
AS621A	6·12 abc	0·012	6·95 ab	0·992*	5·97 abc	−0·106	4·51 ef	−0·534	5·41 bcd	−0·362
Second experiment										
AS616 A	6·10 cde	−1·094*	7·45 ab	0·794*	6·60 bcd	−0·292	5·47 fge	−0·308	7·50 a	0·902*
AS617 A	6·50 bcd	0·440	5·90 def	0·378	6·80 abc	1·042*	3·10 h	−1·544*	5·15 fg	−0·314
AS618 A	6·10 cde	0·192	5·89 def	0·520	4·72 g	−0·886*	4·88 fg	0·388	5·10 fg	−0·212
AS619 A	6·43 bcd	0·674	2·50 i	−2·718*	5·90 def	0·446	6·00 cde	1·660*	5·10 fg	−0·060
AS621 A	6·50 bcd	−0·210	7·20 abc	1·028*	6·10 cde	−0·308	5·10 fg	−0·194	5·80 def	−0·314

[a]Means followed by different letters are significantly different at $P = 0{\cdot}05$ (Newman–Keuls test).
*Significant at $P = 0{\cdot}05$.

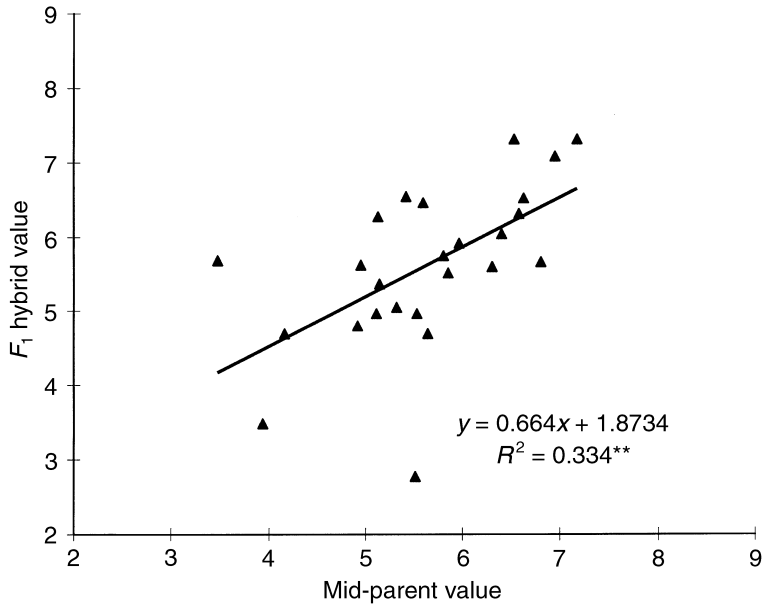

Figure 1 Regression of hybrid value on mid-parent value (means of two experiments) for resistance to *Phoma*. Scale 1–9: resistance to susceptibility.

lines AS618A, AS619A and AS614R (Table 3) indicate their value as combiners for breeding to improve partial resistance to *Phoma*. Both significant positive and negative estimates of GCA and SCA for resistance to *Phoma* were obtained, as has also been shown for resistance of sunflower to other diseases, such as *Phomopsis* (Vranceanu *et al*., 1992) and *Sclerotinia sclerotiorum* (Robert *et al*., 1987; Castano *et al*., 1992).

The variance attributable to male-sterile lines was more important than that of restorers (Table 4), probably because of the existence of either or both maternal effects and more effective genes for resistance in the male-sterile inbred lines used in this experiment. A complete diallel programme should be undertaken to determine the reciprocal effects of restorers and male-steriles in the genetic control of resistance to *Phoma* in sunflower.

This work confirms the hypothesis that resistance to *Phoma* in sunflower is controlled by several genes with additive and dominant effects, and that it can be improved by using inbred lines presenting significant GCA effects for resistance in breeding programmes.

Acknowledgements

The authors thank ASGROW France SA for supporting this work.

References

Acimovic M, 1984. Sunflower diseases in Europe, the United States and Australia. *Helia* 7, 45–54.

Arabi MI, Barrault G, Sarrafi A, Albertini L, 1992. Variation in

the resistance of barley cultivars and the pathogenicity of *Drechslera teres* f.sp. *maculata* and *D. teres* f.sp. *teres* isolates from France. *Plant Pathology* **41**, 180–6.

Brunin B, Lacoste L, 1970. Recherches sur la maladie du colza due à *Leptosphaeria maculans* (Desm.), pouvoir pathogène des ascospores. *Annales de Phytopathologie* **2**, 477–88.

Carson ML, 1991. Relationship between *Phoma* black stem severity and yield losses in hybrid sunflower. *Plant Disease* **75**, 1150–3.

Castano F, Henery-Tardin MC, de Tourvieille Labrouche D, 1992. The inheritance and biochemistry of resistance to *Sclerotinia sclerotiorum* leaf inoculations in sunflower (*Helianthus annuus* L.). *Euphytica* **58**, 209–19.

Donald PA, Venette JR, Gulya TJ, 1987. Relationship between *Phoma macdonaldii and* premature death of sunflower. *Plant Disease* **71**, 466–8.

Falconer DS, 1972. *Introduction to Quantitative Genetics*. London, UK: Olivier and Boyed.

Hua Z, Ma G, 1996. A review of sunflower diseases research of China. In: *Proceedings of the 14th International Sunflower Conference, Beijing, China*. Shenyang, China: Academy of Agricultural Science, 2, 754–9.

Newman PL, Bailey DJ, 1987. Screening for resistance to canker (*Leptosphaeria maculans*) in winter oilseed rape (*Brassica napus* spp. *oleifera*). *Plant Pathology* **36**, 346–54.

Penaud A, 1996. *Phoma* du tournesol: recherche des époques de contamination et mise au point de la protection fongicide. In: *Proceedings of the 14th International Sunflower Conference, Beijing, China*. Shenyang, China: Academy of Agricultural Science, 2, 694–9.

Peres A, Lefol C, 1996. *Phoma macdonaldii* Boerema: élément de biologie et mise au point d'une méthode de contamination artificielle en conditions controlées. In: *Proceedings of the 14th International Sunflower Conference, Beijing, China*. Shenyang, China: Academy of Agricultural Science, 2, 687–93.

Peres A, Allard AM, Deverchère J, Penaud A, 1994. *Phoma* du tournesol: etude de la protection fongicide au champ. In: *4ème Conference Internationale Sur les Maladies Des Plantes, Bordeaux, France*. Paris, France: Association Nationale de Protection des Plantes, 1179–85.

Robert N, Vear F, de Tourvieille Labrouche D, 1987. L'hérédité de la résistance au *Sclerotinia sclerotiorum* (Lib.) de Bary chez le tournesol. I. Etude des réactions à deux tests mycéliens. *Agronomie* **7**, 423–9.

Vranceanu AN, Craiciu DS, Soare G, Pacureanu M, Vainescu G, Sandu I, 1992. Sunflower genetic resistance to *Phomopsis helianthi* attack. In: *Proceedings of the 13th International Sunflower Conference, Pisa, Italy*, 1301–6.

Williams PH, Delwiche PA, 1979. Screening for resistance to blackleg of crucifers in the seedling stage. In: *Proceedings of Eucarpia 1979 Conference, Wageningen, The Netherlands*, 164–70.