Inheritance of Powdery Mildew Resistance in Strawberry Lines from the Israeli Germplasm Collection

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Abstract. Powdery mildew (PM) of strawberry (*Fragaria ×ananassa*), caused by the obligate parasite *Podosphaera aphanis* syn., is a major fungal disease of strawberry crops all over the world. This disease is particularly severe in the humid environments of covered, small tunnels and greenhouses. Marked variation in the susceptibility of different strawberry cultivars to PM and a high heritability of PM resistance have been reported in strawberry breeding populations. This implies a great potential for the breeding of new cultivars with enhanced disease resistance.

Sixty accessions from the Israeli strawberry germplasm collection were screened for relative resistance to PM. Despite high levels of PM inoculation and the high planting density in the greenhouse, we were able to identify improved resistance in some of the examined strawberry accessions. The most resistant and most susceptible accessions were used as parents and hybridized to yield PM-segregating populations. Four parallel crosses were made, and about 120 F1 progeny plants from each combination were screened in four replicates for PM disease severity. We observed an intermediate level of heritability for PM resistance in the different populations and resistance sources, $H^2 = 0.46–0.56$. The results of this study of two different types of populations reveal a strong environmental effect on PM disease severity, even in situations in which there is a high level of inoculation.

Powdery mildew of strawberry (*Fragaria ×ananassa* Duch.), caused by the obligate parasite *Podosphaera aphanis* syn., is one of the major fungal diseases of this crop worldwide (Maas, 1998; Spencer, 1978). This crop-specific pathogen infects all aboveground host organs, including leaves, petioles, stolons, flowers, and fruit. Dense mycelial coverage of the leaf surfaces causes a reduction in photosynthesis, which can lead to necrosis and eventual defoliation (Maas, 1998). Yield losses may also result from infections of flowers and fruit, organs that are susceptible to infection at all stages of their development. Infected green fruit fail to ripen and infected ripe fruit remain soft, have a shortened shelf life, and possess small seeds (Spencer, 1978). Yield losses in strawberry due to PM may reach 60% in certain cultivars (Nelson et al., 1995), emphasizing the importance of cultivar choice as a major component of an effective PM management program. Although there have been no reports of absolute resistance, relative tolerance to the disease has been reported in California (Nelson et al., 1996) and Norway (Davik and Honne, 2005) in certain short-day and day-neutral cultivars (Darrow et al., 1954; Miller and Waldo, 1957; Simpson, 1987).

The heritability of PM resistance in everbearing and day-neutral strawberry seedlings was found to be affected by both general combining ability (GCA) and specific combining ability (SCA) (Simpson, 1987). Cytoplasmic effects have also been thought to be involved in the inheritance of mildew resistance (Harland and King, 1957). When PM resistance was evaluated in 17 strawberry progeny populations under open field and greenhouse conditions (Nelson et al., 1995), high levels of heritability were found for both disease incidence ($H^2 = 0.44–0.71$) and disease severity ($H^2 = 0.70–0.94$). The higher H^2 values were obtained at high infection levels, which are thought to enhance the reliability and repeatability of the screening procedure. The high similarity of the results of field and greenhouse trials (Nelson et al., 1995, 1996) confirms the utility of efficient greenhouse screening for the identification of the most resistant genotypes for breeding programs. Considerable variation in powdery mildew resistance was also reported among 63 *F. ×ananassa* cultivars screened in the field and, in that study, the broad sense heritability (H^2) of this trait was in the range of 0.44 to 0.50 (Davik and Honne, 2005).

In a preliminary screening of the Israeli strawberry germplasm collection for lines exhibiting relative PM resistance, we observed excellent PM resistance in several cultivars under conditions of extreme conidial inoculation. Based on this initial screening, we selected the most resistant and most susceptible genotypes for use as parents for the production of PM-segregating populations to be used for analyses of the heritability of PM resistance.

Materials and Methods

Screening test for PM resistance. PM inoculum was obtained from severely infected plants of the susceptible cultivars Toyonaka and Tamar grown in a greenhouse at the Volcani Center, Bet Dagan, at temperatures of 22 to 25 °C day/15 to 18 °C night. Plants of several cultivars (four plants/cultivar) were cultivated in a soilless potting mix in 13-cm pots and were arranged randomly in high planting density in the greenhouse. Dry PM conidia from the infected leaves were applied to uninfected leaves. Disease symptoms appeared on leaves 7 to 10 d after inoculation and disease severity was evaluated weekly until 35 d postinoculation. Disease severity was rated on a scale of 0 to 100%, according to the extent of disease symptom coverage of infected leaves (Amsalem et al., 2005).

Plant material and breeding populations. The strawberry

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Table 1. List of PM-segregating populations.

Population ID	Susceptible parent	Resistant parent	No. of progeny
PM 14	Tamar	Irvine	120
PM 538	Tamar	Buba	138
	Resistant parent	Resistant parent	
PM 628	Irvine	Line 628	124
PM 214	Irvine	Line 214	123

cultivars tested for relative PM resistance came from the Israeli strawberry germplasm collection of the Volcani Center. The extremely resistant and extremely susceptible cultivars were crossed to produce the PM-segregating populations listed in Table 1. Seedlings of each population were vegetatively propagated to produce four daughter plants, which were arranged randomly in the greenhouse and used for evaluating the effect of the genetic backgrounds on PM severity.

Heritability analysis. The extent of PM mycelia coverage was scored in the four daughter plants generated from each seedling progeny and data were statistically analyzed using JMP 5.1 software (SAS Institute, Cary, NC). The variance components were calculated using a two-factorial model: population and plant replications (random effect) analysis. The broad sense heritability of PM resistance for each population was estimated using the following REML variance components: 1) within replications (σ_w^2), to represent the environmental effect and random error; and 2) between replications (σ_b^2), to represent the variance due to genotype.

$$H_b^2 = \frac{\sigma_b^2}{\sigma_b^2 + \sigma_w^2}$$

Results and Discussion

Evaluating PM severity in the different Israeli cultivars. Sixty accessions from the Israeli strawberry collection were screened for relative resistance to PM in two parallel experiments. Despite the high level of PM inoculation and the high planting density in the greenhouse, good resistance was identified in some of the strawberry accessions, as shown in Fig. 1. Relative to the susceptible cultivars, the resistant lines showed either no symptoms or only very minor, localized PM patches. From these genetic resources, we identified extremely resistant and susceptible cultivars for use as parents in the development of PM resistant/susceptible segregating populations. The progress of PM infection on strawberry leaves through 5 weeks after inoculation is shown in Fig. 2. Strawberry Tamar was chosen for use as susceptible parent and accessions 214 and 628, as well as cvs. Buba and Irvine (data not shown), were selected for use as resistant parents in the crosses that are listed in Table 1.

Development of PM-segregating populations. In order to establish a strawberry population that would segregate for PM resistance, four parallel crosses were made (Table 1) and about 125 F1 progeny plants from each cross were grown in trays under shade. To distinguish between one and two resistance donors in the segregating populations, two different crosses were made: populations PM214 and PM628, which combined two resistant parents, and populations PM14 and PM538, which combined resistant and susceptible parents. PM disease severity was evaluated in each individual seedling of the segregating populations (four vegetative replications/seedling) 1 month after a natural PM epidemic began. These disease severity data were then used for analysis of the heritability of resistance.

Distribution of PM in the segregating populations. The average score for PM severity in populations PM 628 and PM 214 was about 30%, which is relatively low compared to the levels observed in populations PM 14 and PM 538, which had more then 40% mycelium coverage (Table 2). Within the populations PM 14 and PM 538, there were many progeny with very severe disease, which in some cases reached a maximum level of 90% mycelium coverage. In comparison, the maximum disease severity in populations PM 628 and PM 214 was 60%.

The observed disease distributions demonstrate a positive correlation between the population mean (Table 2) and the total variance (Table 3). The populations with higher levels of disease also have higher levels of variation in PM severity; this is also known as a scaling effect (Falconer, 1960) Although mildew infections were more consistent and mild in populations PM214 and PM628, the relative component of the environment variance (within replications) and the experimental error remained about 50%. These results, from two different population types, reveal that PM severity is strongly affected by environmental conditions.

Heritability of PM resistance. The broad sense heritability

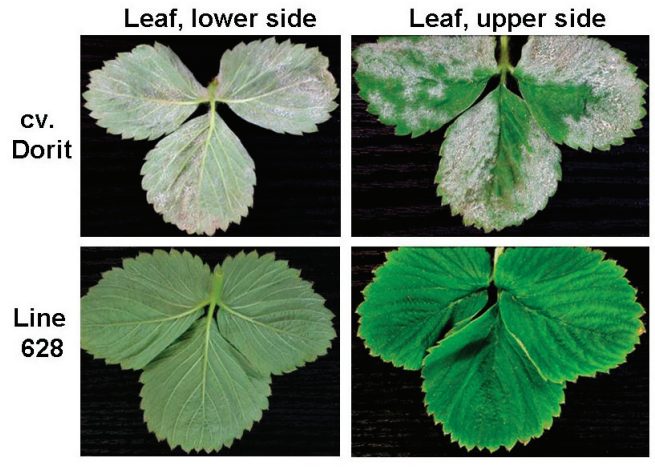


Fig. 1. Powdery mildew on leaves of the susceptible strawberry cultivar Dorit and the resistant line 628.

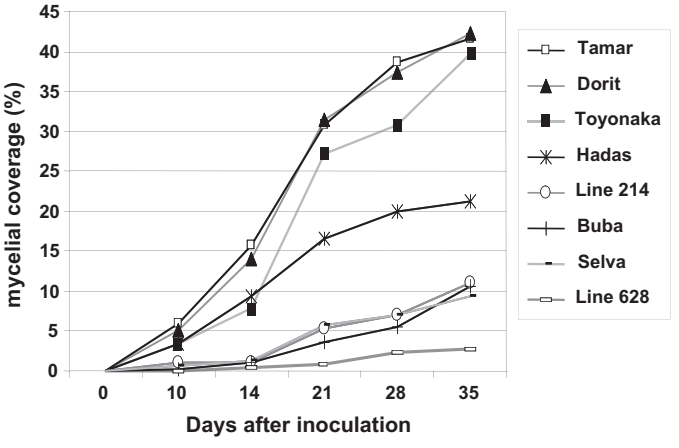


Fig. 2. The progress of PM infection of strawberry leaves 0–5 weeks after inoculation. Values are means of PM symptoms coverage from five strawberry plants.

Table 2. Powdery mildew disease severity (percentage of leaf surface covered by mycelia) means and the standard errors of the means (SE) in the PM-segregating populations.

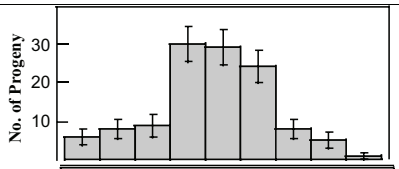
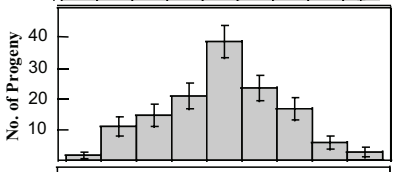
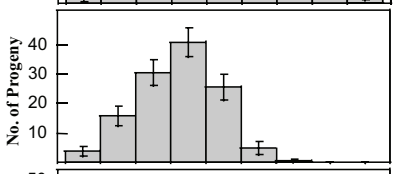
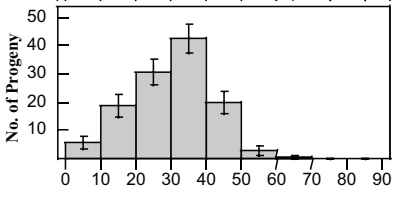
Population ID	Distribution	Mean	SE
PM 14		41.38	1.48
PM 538		43.69	1.49
PM 628		30.82	1.04
PM 214		29.44	1.09

Table 3. Results from the analysis of the variance (variance analysis) of powdery mildew severity in the different populations, the variance components between and within replications and the estimation of the heritability of resistance.

Population ID	Variance component	Variance level	H^2
PM 14	Between replications	216.04	0.54
	Within replications	181.011	
PM 538	Between replications	253.63	0.56
	Within replications	196.34	
PM 628	Between replications	110.88	0.53
	Within replications	98.47	
PM 214	Between replications	112.26	0.46
	Within replications	133.78	

of PM resistance observed in this experiment is estimated to be about 0.5 in all four populations (Table 3). In similar studies by Nelson et al. (1995), the heritability of mildew resistance was estimated to be between 0.44 and 0.71. Our heritability estimate for PM resistance is also in agreement with a recent publication by Davik and Honne (2005).

PM resistance in strawberry seems to be a complex trait that could be affected by several loci and gene interactions, as has been observed in studies of mildew resistance in apple (James et al., 2004). Future work aimed at identifying the specific loci involved in resistance and the development of molecular markers for these loci could greatly simplify the breeding of strawberry cultivars with durable mildew resistance.

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