Theor Appl Genet (2003) 107:256–262
DOI 10.1007/s00122-003-1240-1

M. Linde · Th. Debener

Isolation and identification of eight races of powdery mildew of roses (*Podosphaera pannosa*) (Wallr.: Fr.) de Bary and the genetic analysis of the resistance gene *Rpp1*

Received: 15 June 2002 / Accepted: 14 November 2002 / Published online: 19 March 2003

Abstract Powdery mildew, caused by *Podosphaera pannosa*, is one of the most-severe diseases of roses grown under glass. The differentiation into physiological races and the genetic analysis of resistance in a segregating host population was investigated using single conidial isolates of the pathogen. Using ten rose genotypes, all eight isolates of the pathogen could be ascribed to different races. Five races were isolated from one location, which indicates that populations of *P. pannosa* exhibit a high racial diversity. Infection experiments in a backcross-population of 114 rose plants resulted in a 1:1 segregation, suggesting control by a single dominant gene. *Rpp1* is the first resistance gene against rose powdery mildew to be described.

Keywords *Rosa* · *Sphaerotheca pannosa* · Fungal races · Plant resistance · Genetic analysis

Introduction

Powdery mildew fungi are ubiquitous pathogens of various field and greenhouse crops worldwide. Because of the constant favourable environment in greenhouse cultures, they are extremely aggressive pathogens. The losses in the production caused by powdery mildews are difficult to estimate; however, the cost for their control reaches about \$6,000 CN ha^{-1} per year for the three most-important greenhouse crops: cucumber, tomato and roses (Paulitz and Belanger 2001) in Canada alone. Powdery mildew of roses was first described by Theophrastus in 300 BC (Horst 1983). It is caused by the obligate biotrophe *Podosphaera pannosa* (Wallr.: Fr.) de Bary, an ascomycete, which had long been known as *Sphaerotheca pannosa* var. *rosae* (Wallr.: Fr.) Lév (Braun and Takamatsu 2000).

Whereas black spot is considered to be the most severe disease of field-grown roses (Horst 1983; Debener et al. 1998; Malek von and Debener 1998; Yokoya et al. 2000), powdery mildew is the major fungal pathogen of roses grown in greenhouses (Horst 1983; Linde and Shishkoff 2003). There have been many reports on the development, epidemiology and host – parasite relationship of *P. pannosa* in which the environmental and cytochemical aspects of the infection process and the defence reaction of the rose were described. Weinhold (1961a, b) concentrated his research on the development of *S. pannosa* on peaches in the field and on germination experiments in the laboratory. He showed the germination of conidia over a wide temperature range from 4 to 36 °C within 24 h, with an optimum between 21 and 27 °C. The length of the latent period of rose powdery mildew ranges from 4 to 10 days within a temperature gradient from 10 to 28 °C and an optimum at 22 °C (Xu 1999). Price (1970) confirmed the temperature requirements from Weinhold and demonstrated the rare occurrence of cleistocarps and the perennation of *P. pannosa* in buds. In examinations between September 1966 and December 1968 he found cleistocarps on only 37 out of 741 genotypes. Mence and Hildebrandt (1966) and Conti et al. (1985) concluded that morphological barriers only play a minor role for resistance, if any at all, and do not cause poor conidial germination. One year later Conti et al. (1986) suggested the deposition of phenolic compounds at the penetration side as one factor of rose resistance to powdery mildew. Ferrero et al. (2001a) observed a lower cuticle transpiration rate for one resistant genotype compared to one susceptible genotype. Development of powdery mildew seems to be retarded by water droplets caused by spraying in the first 6 h after inoculation, probably due to disturbances within the initial penetration phase (Perera and Wheeler 1975). In ultrastructure examinations, Hajlaoui et al. (1991) showed the formation of multilobed haustoria with an extrahaustorial matrix free of chitin, pectin or cellulose. The formation of papillae at the

Communicated by F. Salamini

M. Linde (✉) · T. Debener
Federal Centre for Breeding Research on Cultivated Plants,
Institute for Ornamental Plant Breeding, Bornkampsweg 31,
D-22926 Ahrensburg, Germany,
e-mail: M.Linde@bafz.de
Fax: +49-4102-51124

penetration site was often associated with a restricted growth of the penetration peg. Hajlaoui also demonstrated the formation of a cellulose-rich collar around the haustorial neck of *P. pannosa*. Many reports considered the evaluation of powdery mildew resistant rose species, varieties and hybrids (Ferrero et al. 2001b). Mence and Hildebrandt (1966) tested 17 cultivated varieties and six species with an inoculum from two sources with a detached leaflet technique. Conidia from *Rosa virginiana* readily infected *Rosa rugosa* but did not infect most of the varieties. Atkiss (1978) presented a list of more than 30 disease resistant varieties revealed by the evaluation of 11 public rose gardens in the U.S.A. over several years. However, up to now there is only limited information about the pathogenic specialization of *P. pannosa*. In 1952, Yarwood reported the existence of two different strains of *P. pannosa* on *Prunus ameniaca* L. fruits. Mence and Hildebrandt (1966) also suggested the existence of two separate forms of the fungus, differing in host range and size of conidia. In contrast to these authors, which used mixed inoculum, Bender and Coyier (1984) and Leus et al. (2002) established monoconidial isolates. Bender and Coyier identified five races of *P. pannosa* with a differential set of four rose cultivars out of nine isolates sampled in Oregon (U.S.A.). Leus et al. couldn't distinguish the eight isolates from Belgium with their differential set. Inoculation experiments by these two authors were made by dusting conidia from infected shoots over the test cultivars or by brushing inoculum onto the test plants. No attention was paid to differences in density of the inoculated conidia in the experiments. Here we present data on the isolation and characterisation of physiological races of *P. pannosa* and the first genetic analysis of resistance to the pathogen in a segregating host population.

Materials and methods

Fungal isolates

Eight monoconidial isolates of *P. pannosa* were obtained from naturally infected field plants from four sites (Table 1). Isolates 2, 3, 4, 6 and 9 were obtained from different host genotypes from field plots of the Institute for Ornamental Plant Breeding (Federal Centre for Breeding Research on Cultivated Plants) in Ahrensburg, Germany. Isolate 12, from the rose 'Rebell Kordana' was sampled at Risø National Laboratory in Roskilde, Denmark. Isolates nos. 8 and 10 were collected from two unlabelled cultivars in private gardens and parks in Sarstedt (Germany) and Tel Aviv (Israel). Single powdery mildew conidia from naturally infected leaves were transferred to in vitro cultures of the susceptible genotypes 'Queen Elizabeth' and *94/103-02* using a thin infection needle (0.2 mm). After about 10 days, single conidia or single conidiophores from the established mildew colonies were transferred to new in vitro plants. To eliminate possible contaminations by inadvertent transfer of different conidiophores from one colony, this procedure was repeated five times. The isolates were then considered to be genetically uniform. In order to preserve and multiply the isolates, new in vitro shoots were infected every 3 to 4 weeks by gently rubbing them with infected plants under a clean bench. All in vitro plants were grown in 250-ml glass vessels at 23 to 24 °C inside the vessel, and in a light cabinet (16-h light, approximately 1,500 cd sr/m^2) on culture media modified after Murashige and Skoog (1962) described by Dohm et al. (2001).

Plant cultivars

The following rose genotypes were used for testing resistance to *P. pannosa*: *Rosa multiflora* Thunb. 27-02, *R. multiflora* Thunb. 27-04, and *Rosa wichuraiana* Crépin 99/01, 'Caramba', 'Elina', 'Heckenzauber', 'Pariser Charme', 'Queen Elizabeth', 'Rebell Kordana'. Rose genotypes 82/78-1, 81/42-15, 88/124-46, 91/100-5, 93/1-117, 93/1-119, 95/13-39, 95/13-79 and 95/13-90 were derived from *R. multiflora* crossings made in the Institute for Ornamental Plant Breeding. These plants were grown mildew-free in a climate chamber (15-h light: 25 °C, 75% rh; 9-h dark: 18 °C, 75% rh) in 3- or 5-l plastic pots in rose substrate with pH 5.5 [70 vol% white peat, 30 vol% coco fibre, 1.2 kg/m^3 of PG-Mix NPK fertilizer and 100 g/m^3 of trace-element fertilizer (Jost, Iserlohn)]. For the genetic analysis of the resistance to race 9, the following plant material was used: in 1995 the susceptible diploid pollen donor 82/78-1 was crossed to the resistant diploid line 88/124-46. The resulting resistant genotype 95/13-90 was backcrossed in 1996 to the susceptible 82/78-1, resulting in the population 97/9. Plants were grown in a greenhouse in rose substrate and sprayed at regular intervals with non-systemic fungicides to prevent powdery mildew contamination. Young pesticide-free leaves from these plants were sampled only from November to May when the powdery mildew pressure from outside was very low.

Inoculation assay

Two different inoculation towers were used in two types of experiments: six Petri dishes with the leaflets of all 18 rose genotypes of the genotype assortment were placed under an inoculation tower, constructed from two metal-barrels (diameter 28.5 cm, height 90 cm) and a funnel. For the genetic analysis of resistance a larger inoculation tower, made from a metal-barrel (diameter 51 cm, height 98 cm) and a funnel was used. With this set-up leaflets from 78 genotypes of the population 97/9 were inoculated in one experiment. For both types of experiment six to nine leaflets each (3rd–5th unfolded leaves from the shoot tip) from three rose genotypes were placed in glass Petri dishes on water-agar (0.5% agar) with 0.03% benzimidazole to prevent fungal contam-

Table 1 Origin of the monoconidial isolates

Isolate	Country	Town	Host genotype[a]	Host genotype[b]
2	D	Ahrensburg	'Caramba'	'Queen Elizabeth'
3	D	Ahrensburg	*R. caudata*	94/103-02
4	D	Ahrensburg	'Sommerwind'	'Queen Elizabeth'
6	D	Ahrensburg	'Superstar'	'Queen Elizabeth'
8	D	Sarstedt	Unknown	94/103-02
9	D	Ahrensburg	97/7-13	94/103-02
10	IL	Tel Aviv	Unknown	'Queen Elizabeth'
12	DK	Risø	'Rebell Kordana'	'Queen Elizabeth'

[a] Rose genotype from which the conidia for the isolate were derived
[b] In-vitro rose genotype on which the isolates were maintained

Table 2 Sporulation data for eight isolates of *P. pannosa* on 18 rose genotypes

Rose genotypes	Single conidial isolates															
	2		3		4		6		8		9		10		12	
	m[a]	se[b]	m	se	m	se	m	se	m	se	m	se	m	se	m	se
91/100-5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
95/13-90	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
93/1-117	0	0	0	0	0	0	0	0	0	0	50	10	0	0	0	0
88/124-46	0	0	0	0	0	0	0	0	0	0	0	0	4	9	0	0
'Heckenzauber'	3	6	0	0	0	0	0	0	8	13	0	0	0	0	0	0
95/13-79	0	0	0	0	0	0	17	9	0	0	2	3	0	0	0	0
95/13-39	0	0	0	0	0	0	0	0	1	2	2	3	0	0	0	0
Sp.-18 Scharberg	0	0	0	0	0	0	2	4	4	6	0	0	4	9	0	0
R. wichuraiana 99/01.	0	0	0	0	0	0	3	3	0	0	2	3	10	14	0	0
R. multiflora 27-02	0	0	3	6	0	0	0	0	4	6	2	3	2	5	0	0
'Caramba'	7	12	0	0	3	3	7	13	10	10	0	0	1	2	4	4
'Queen Elizabeth'	20	10	0	0	9	8	30	10	24	11	0	0	22	8	40	14
'Elina'	20	10	0	0	11	6	44	14	22	13	0	0	2	5	3	3
'Rebell Kordana'	23	6	0	0	4	5	26	11	8	8	0	0	18	15	1	2
82/78-1	2	3	0	0	9	9	20	7	16	11	17	6	18	8	2	3
93/1-119	43	6	0	0	40	14	32	5	24	15	53	12	32	13	50	13
R. multiflora 27-04	2	3	0	0	4	5	18	9	16	9	12	8	16	9	3	4
'Pariser Charme'	3	3	0	0	4	5	26	14	18	13	10	9	22	8	9	6

[a] Arithmetical mean of 3–5 replications
[b] Standard error of the mean

ination. About two conidia/mm^2 were dispersed over the leaves, by placing infected in vitro shoots into the opening of the funnel and blowing the conidia with compressed air (approximately 200 kPa) into the tower. To estimate the density of conidia, three microscope slides were placed on different positions between the Petri dishes under the inoculation tower. The number of viable, colourless and undamaged spores per field of vision was counted under the microscope at 100-fold magnification. The median number of conidia/mm^2 of 12 counts per slide was used to calculate the mean for the three slides. This served to estimate the conidial density on the leaf material. The Petri dishes were incubated in a plant growth chamber for 10 days (16-h light, 22 °C).

Disease assessment

Ten days after inoculation, percentages of the leaf areas covered with conidiophores were estimated in 10% steps (from zero to 100%) with a stereomicroscope (8–50-fold magnification) and used as a disease index (DI). Genotypes with only single conidiophores on the whole leaf material in one experiment were scored as having a DI of 5%. Rose genotypes showing a DI of 10% or more were considered to be susceptible. Mean DI values of lower than 5% and with no single estimation over 5% were treated as resistant. For the characterisation of the isolates, the mean and the standard error of 3–5 replicates were calculated for each rose genotype in combination with each isolate (Table 2). For the genetic analysis of resistance in the population 97/9, the inoculations with isolate 9 were repeated five to eight times for 114 individuals. The minimum and maximum values were excluded and the mean was calculated.

Results

Inoculation assay

Leaflets, placed on benzimidazole-agar, normally stayed green and healthy even after incubation periods of 10 days in a light thermostat. Infections with *Botrytis cinerea* were only rarely observed (data not shown). Leaflets with strong *Botrytis* infections were excluded from the evaluation. The distribution of the conidia over the area under the inoculation tower was relatively regular, with differences below 15% between the mean conidial densities of the three microscope slides in one inoculation experiment. Between the 50 different inoculation experiments the average conidial density, as the mean of the counts of three microscope slides each, varied between 1.7 and 2.4 conidia/mm^2.

Characterisation of monoconidial isolates of *P. pannosa*

From the 18 rose genotypes of the differential assortment, 16 showed the production of conidiophores for at least one isolate of *P. pannosa* (Table 2). Only the genotypes 91/100-5 and 95/13-90 remaind without visible symptoms for all isolates. With a mean of 34% leaf area covered with conidiophores, the genotype 93/1-119 (Fig. 1) showed the highest susceptibility of all tested cultivars. All eight single conidial isolates could be distinguished with a subset of ten rose genotypes out of the 18 investigated (Table 3). These genotypes displayed varying degrees of susceptibility to the different isolates. Genotype 95/13-79 was only infected by isolate 6, 'Pariser Charme' was resistant to the monoconidial lines 2–4, whereas genotype 93/1-119 was highly susceptible for all isolates except isolate 3. Of the five isolates collected in the small area at the Institute for Ornamental Plant Breeding in Ahrensburg, any two could be distinguished by at least three differential reactions. Due to these clear differences between compatible and incompatible interactions with the host genotypes the isolates can be classified as different physiological races of *P. pannosa*.

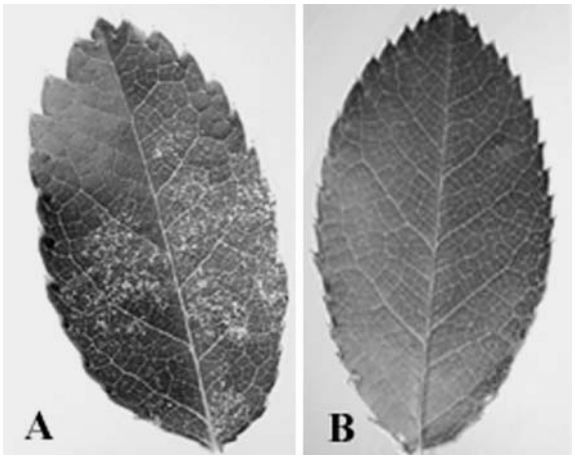

Fig. 1A, B Leaf of the susceptible genotype 93/1-119 (**A**) with about 60% leaf area covered with conidiophores 10 days after inoculation. The resistant plant 95/13-90 (**B**) shows no infection

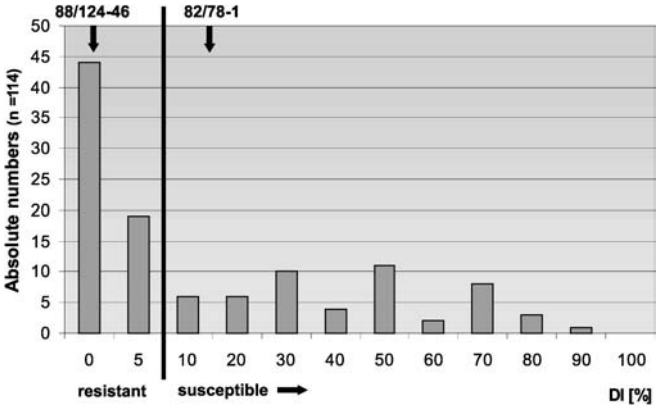

Fig. 2 Frequency distribution of the disease index (DI) in the BC population 97/9 inoculated with powdery mildew race 9. Disease index: percentage leaf area covered with conidiophores 10 days after inoculation (mean of 5–8 replications)

Table 3 Resistance of the various rose genotypes to single conidial isolates from locations identified in Table 1

Rose genotypes	Single conidial isolates							
	3	12	4	2	9	10	8	6
93/1-117	0[a]	0	0	0	1[b]	0	0	0
95/13-79	0	0	0	0	0	0	0	1
'Caramba'	0	0	0	1	0	0	1	1
'Rebell Kordana'	0	0	0	1	0	1	1	1
'Elina'	0	0	1	1	0	0	1	1
R. multiflora 2704	0	0	0	0	1	1	1	1
82/78-1	0	0	1	0	1	1	1	1
'Pariser Charme'	0	1	0	0	1	1	1	1
'Queen Elizabeth'	0	1	1	1	0	1	1	1
93/1-119	0	1	1	1	1	1	1	1

[a] 0 indicates resistance
[b] 1 indicates susceptible reaction; mean of 3–5 repeated experiments

Segregation analysis

To gain an insight into the genetic basis of the resistance of roses to powdery mildew a diploid population (97/9) was analysed with race 9 in repeated inoculation assays. The population 97/9 is derived from a cross between the resistant 88/124-46 and the susceptible male parent 82/78-1, backcrossed to 82/78-1. It was chosen for genetic analysis because in previous investigations a single dominant gene for blackspot resistance was already mapped in this population relative to the molecular markers. The isolate number 9 was chosen because it was the first isolate identified that distinguished the parental genotypes. The susceptible recurrent parent 82/78-1 displayed a DI of 15%. In contrast, the resistant parents 88/124-46 and 95/13-90 showed no production of conidiophores in any experiment. The 114 plants of the backcross population 97/9 displayed a wide range of susceptibility against physiological race 9 (Fig. 2). Apart from plants without any visible symptoms the group of plants with a disease index of 5% was also considered as being resistant, because it comprised plants with only occasional occurrence of single conidiophores in a fraction of the experiments (see Materials and methods). Sixty three genotypes were scored as resistant, from which 44 had a DI of zero, 19 genotypes had a mean DI of 5% and the remaining 51 susceptible genotypes showed a DI between 10 and 90%. The DI values of the segregating population greatly exceeded the mean DI of the susceptible 82/78-1 (DI = 15%). With 63 resistant versus 51 susceptible plants, the population displayed a 1:1 segregation ($\chi^2 = 0.913$, $P = 0.340$) as expected for the control by a single dominant resistance gene in a backcross. The segregation for resistance to the powdery mildew race 9 in the population 97/9 differs significantly from the data obtained for the black-spot isolate DortE4 in the same population. Within 85 genotypes scored for both pathogens we found 30 recombinant plants. So, there is strong evidence for two unlinked loci for these two resistance genes in roses.

Discussion

The results of the present study confirm and extend the identification of the physiological races of *P. pannosa* by Bender and Coyier (1984). With a set of ten different rose genotypes, all eight tested isolates from diverse host genotypes could be identified as different races. It is highly probable that the observed pattern follows the 'gene-for-gene system' since a so-called "quadratic check" for different interactions is possible (Table 4).

Table 4 Quadratic check for the interaction of races 2 and 10 and two rose genotypes indicating a gene for gene system

Genotype	Race 2	Race 10
R. multiflora 2704	0	1
'Elina'	1	0

This would be in line with several other powdery mildew pathosystems for which gene-for-gene relationships have been postulated (Geiger and Heun 1989; Jorgensen 1994; Keller et al. 2000).

Five races collected from different rose genotypes in the field plots of the Institute for Ornamental Plant Breeding, could also be clearly differentiated with the ten rose genotypes. This high diversity in the pathogen population could be explained by the large number of different rose cultivars, wild species and cross-bred progenies in the Ahrensburg area, containing various resistance genes. According to the gene-for-gene hypothesis (Flor 1942) the composition of avirulence genes in a pathogen population is dependent on the diversity of the resistance genes in the host (Bousset et al. 2002). Comparable high racial diversities were observed in different other species of powdery mildew pathosystems. With a differential set of 50 groundsel inbred lines all 24 collected single-conidial-chain isolates of *Erysiphe fischeri*, the cause of powdery mildew on *Senecio vulgaris*, from two UK sites could be identified as different virulence phenotypes, suggesting also an enormously variable pathosystem (Bevan et al. 1993). In the well-described system *Erysiphe graminis* f. sp. *hordei* on barley, 56 and 86 pathotypes were recorded in three regions of northern France in a 5-year period with two differential sets respectively (Andrivon and De Vallavieille-Pope 1993). For the differentiation, the authors collected 721 and 510 individuals during the flowering seasons of the host using a spore trap on a car roof. In a review on population genetics in barley mildew, Wolfe and McDermott (1994) presented data on 9,000 isolates sampled all over Europe from which 609 virulence phenotypes could be identified. An analysis of 363 isolates sampled from a single field in Switzerland already revealed 111 pathotypes. From these observations the authors concluded that *E. graminis* displays an extremely high genetic diversity even in small local populations. From two mildew populations on wild barley in Turkey, Braun and Turgut (1995) isolated 31 and 61 pathotypes respectively, also demonstrating a high genetic diversity of mildew populations on wild plant species.

Yarwood (1952) and Mence and Hildebrandt (1966) suggested the existence of two different strains of *P. pannosa* from inoculation experiments with naturally infected leaves of roses, not using single conidial isolates which are essential for characterising physiological races of plant pathogens (Vanderplank 1978). Five races of *S. pannosa* out of nine isolates from two sites in the U.S.A. were characterised by Bender and Coyier (1984) with a differential set of five cultivars. Leus et al. (2002) could not distinguish any pathotypes from eight isolates sampled at five places in Belgium with a set of seven rose genotypes. In these investigations the infections were made by dusting conidia from infected shoots over the test cultivars or by brushing inoculum onto the test plants, not really securing the inoculation of the leaf material with a similar conidia density in the repeated experiments. As a quantitative trait, the severity of the infection is possibly determined by several minor genes and, therefore, the results could depend highly on the inoculation conditions, especially on the conidial density. In this investigation, the infections were carried out in an inoculation tower, resulting in a relatively homogeneous distribution of the conidia over the leaf material (<15% differences between the mean conidial densities of the three microscope slides) of 18 and 78 genotypes respectively. Differences in the estimated disease indices between the replications are probably due to the different age of the sampled leaf material. This error was reduced by repeating the inoculations for each isolate three to five times. To characterise the isolates a large set of 18 different rose genotypes was used in the tests, which could be reduced to ten in the differential assortment. The use of small differential sets could lead to an underestimation of the racial diversity within the pathogen populations, because of the possibly low number of resistance genes present in these genotypes. Leus et al. (2002) used a set of five commercial cultivars, which were infected by all isolates, and two species, from which *Rosa laevigata anemoides* was also susceptible for all isolates and *R. wichuraiana* showed only minor infections for two isolates. Only two races of *Sphaerotheca fusca* on cucurbits were found within 41 isolates from 30 locations in Crete (Vakalounakis and Klironomou 1995). They used a differential set of two genotypes resistant to all isolates, one generally susceptible cultivar and only one genotype susceptible to 18 of the 41 isolates.

For the analysis of the inheritance of a powdery mildew resistance gene in roses we looked at the segregation for resistance to race 9 in a diploid backcross population of 114 genotypes. With 63 resistant versus 51 susceptible plants, the BC population exhibits a clear 1:1 segregation as expected for the control by a single dominant resistance gene. Thus, for the first time the action of a single dominant resistance gene against *P. pannosa*, called *Rpp1*, was shown by a genetic analysis in roses. Dominance of the resistance gene *Rpp1* could be expected because of the resistance of the F_1 genotype 95/13-90. With DI values between 0 and 90%, the segregating population shows a transgressive segregation clearly exceeding the mean DI of the susceptible 82/78-1 (DI = 15%). This pattern indicates that probably both the resistant female parent and the susceptible pollen donor contribute additional minor alleles with 'negative' effects to the progeny increasing the susceptibility to race 9. Similar examples for transgressive segregation in both directions were found for resistance in wheat against powdery mildew, showing that heterozygous hosts are not necessarily more-resistant to pathogens than homozygous plants (Geiger and Heun 1989). In most QTL studies on plant disease resistance, loci with either positive or negative effects were detected in both parental plants, leading to an improved or decreased resistance in the progeny. In about a third of the studies a large amount of the explained phenotypic variability (25–90%) was caused by one major QTL, modified by the environment or by minor genes (Keller et al. 2000).

From the present study, it can be concluded that the racial structure of *P. pannosa* populations is highly diverse. The fungus seems to exist as a large number of different races. To obtain durable resistance in rose varieties, several resistance genes should simultaneously be introduced to the narrow gene pool of modern roses (Gudin 2000). This can be aided by the use of molecular markers closely linked to known resistance genes like *Rpp1*. First experiments using a bulked segregant strategy with AFLP markers have already been started. The differential set comprises several genotypes, which have been used to generate large segregating populations (e.g. 93/1-117 × 93/1-119, *R. multiflora* 27-04 × *R. wichuraiana*). More than 20 combinations of populations and powdery mildew races provide interesting resources for further genetic analyses in the rose powdery mildew pathosystem. Existing marker maps (Debener and Mattiesch 1999) in some of these populations (e.g. 93/1-117 × 93/1-119) will facilitate molecular mapping of resistance genes in the near future.

Furthermore, the optimised inoculation system can be used to screen commercial cultivars for resistance very effectively, using not only single conidial isolates but also complex racial mixtures, therefore significantly improving the breeding process.

Acknowledgements This project was supported by a grant of the Ministry of Economics, Technology and Transport of the Land Schleswig-Holstein, and by the companies W. Kordes Söhne (Germany) and Rosen Tantau (Germany). We thank K. Sabin for valuable technical assistance and Prof. A. Roberts (UEL, London) for critically reading the manuscript.

References

Andrivon D, De Vallavieille-Pope C (1993) Racial diversity and complexity in regional populations of *Erysiphe graminis* f.sp. *horde* i in France over a 5-year period. Plant Pathol 42:443–464

Atkiss L (1978) Disease resistant rose varieties. American rose annual, pp 99–104

Bender CL, Coyier DL (1984) Isolation and identification of races of *Sphaerotheca pannosa* var. *rosae* Phythopathology 74:100–103

Bevan JR, Crute IR, Clarke DD (1993) Variation for virulence in *Erysiphe fischeri* from *Senecio vulgaris*. Plant Pathol 42:622–635

Bousset L, Hovmøller MS, Caffier V, de Vallavielle-Pope C, Østergård H (2002) Observed and predicted changes over 8 years in the frequency of barley powdery mildew avirulent to spring barley in France and Denmark. Plant Pathol 51:33–44

Braun PW, Turgut I (1995) The virulence structure of mildew populations on wild barley in Turkey. J Plant Dis Protection 102:593–598

Braun U, Takamatsu S (2000) Phylogeny of *Erysiphe*, *Microsphaera*, *Uncinula* (*Erysipheae*) and *Cystotheca*, *Podosphaera*, *Sphaerotheca* (*Cystotheceae*) inferred from rDNA ITS sequences – some taxonomic consequences. Schlechtendalia 4:1–33

Conti GG, Bassi M, Maffi D, Bonecchi R (1985) Host-parasite relationship in a susceptible and a resistant rose cultivar inoculated with *Sphaerotheca pannosa*. I. Fungal growth, mechanical barriers and Hypersensitive Response. Phythopathology 113:71–80

Conti GG, Bassi M, Maffi D, Bocci AM (1986) Host-parasite relationship in a susceptible and a resistant rose cultivar inoculated with *Sphaerotheca pannosa*. II. Deposition rates of callose, lignin and phenolics in infected or wounded cells and their possible role in resistance. Phythopathology 117:312–320

Debener T, Drewez-Alvarez R, Rockstroh K (1998) Identification of five physiological races of black spot, *Diplocarpon rosae*, Wolf on roses. Plant Breed 117:267–270

Debener T, Mattiesch L (1999) Construction of a genetic linkage map for roses using RAPD and AFLP markers. Theor Appl Genet 99:891–899

Dohm A, Ludwig C, Nehring K, Debener T (2001) Somatic embryogenesis in roses. Acta Hort 547:341–348

Ferrero F, Cadour-Marvaldi P, Jacob Y, Coudret A, Sallanon H, Urban L (2001a) Evaluation of the resistance to powdery mildew, *Sphaerotheca pannosa* var. *rosae*, of rose-tree species and hybrids. A. Biological test on excised leaflets, microscopic observations of the fungus, physical evaluation of the leaf cuticle protection. Acta Hort 547:379–381

Ferrero F, Cadour-Marvaldi P, Guiloteau E, Jacob Y, Coudret A, Sallanon H (2001b) Evaluation of the resistance to powdery mildew, *Sphaerotheca pannosa* var. *rosae*, of rose-tree species and hybrids. B. First exploitation of the resistance biodiversity in a crossing program on resistance. Acta Hort 547:379–381

Flor HH (1942) Inheritance of pathogenicity in *Melampsora lini*. Phythopathology 32:653–669

Geiger HH, Heun M (1989) Genetics of quantitative resistance to fungal diseases. Annu Rev Phytopathol 27:317–341

Gudin S (2000) Rose: genetics and breeding. Plant Breed Rev 17:159–189

Hajlaoui MR, Benhamou N, Bélanger RR (1991) Cytochemical aspects of fungal penetration, haustorium formation and interfacial material in rose leaves infected by *Sphaerotheca pannosa* var. *rosae*. Phys Mol Plant Pathology 39:341–355

Horst RK (1983) Compendium of rose diseases. American Phytopathol Soc, St Paul

Jorgensen (1994) Genetics of powdery mildew resistance in barley. Crit Rev Plant Sci 13:79–119

Keller B, Feuillet C, Messmer M (2000) Genetics of disease resistance: basic concepts and application in resistance breeding. In: Slusarenko AJ, Fraser RSS, van Loon LC (eds) Mechanisms of resistance to plant diseases. Kluwer Acad. Publ., Dordrecht Boston London, pp 101–136

Leus L, Van Huylenbroeck J, Van Bockstaele E (2002) Powdery mildew on roses: Pathotype screening. In: Van Huylenbroeck J, Van Bockstaele E, Debergh P (eds) Proc. XX EUCARPIA Symposium on New Ornamentals II. Acta Hort 572:91–95

Linde M, Shishkoff N (2003) Fungi; powdery mildew. In: Roberts A, Gudin S, Debener T (eds) Encyclopedia of rose science. Academic Press Ltd, London (in press)

Malek von B, Debener T (1998) Genetic analysis of resistance to black spot (*Diplocarpon rosae*) in tetraploid roses. Theor Appl Genet 96:228–231

Mence MJ, Hildebrandt AC (1966) Resistance to powdery mildew in rose. Ann Appl Biol 58:309–320

Murashige T, Skoog F (1962) A revised medium for rapid growth and bio assays with tobacco tissue cultures. Physiol Plant 15:473–497

Paulitz CT, Bélanger RR (2001) Biological control in greenhouse systems. Annu Rev Phytopathol 39:103–133

Perera RG, Wheeler BEJ (1975) Effect of water droplets on the development of *Sphaerotheca pannosa* on rose leaves. Trans Br Mycol Soc 64:313–319

Price TV (1970) Epidemiology and control of powdery mildew (*Sphaerotheca pannosa*) on roses. Ann Appl Biol 65:231–248

Vakalounakis DJ, Klironomou E (1995) Race and mating type identification of powdery mildew on cucurbits in Greece. Plant Pathal 44:1033–1038

Vanderplank JE (1978) Genetic and molecular basis of plant pathogenesis. Springer Verlag, Berlin Heidelberg New York

Weinhold AR (1961a) The orchard development of peach powdery mildew. Phythopathology 51:478–481

Weinhold AR (1961b) Temperature and moisture requirements for germination of conidia of *Sphaerotheca pannosa* from peach. Phythopathology 51:699–703

Wolfe MS, McDermott JM (1994) Population genetics of plant pathogen interactions: the example of the *Erysiphe graminis-Hordeum vulgare* pathosystem. Annu Rev Phytopathol 32:89–113

Xu X-M (1999) Effects of temperature on the latent period of the rose powdery mildew pathogen, *Sphaerotheca pannosa*. Plant Pathol 48:662–667

Yarwood CE (1952) Apricot powdery mildew from rose and peach. Calif Dept Agric Bull 41:19–25

Yokoya K, Kandasamy KI, Walker S, Mandegaran Z, Roberts AV (2000) Resistance of roses to pathotypes of *Diplocarpon rosae*. Ann Appl Biol 136:15–20