



RESEARCH ARTICLE

Evaluation for powdery mildew resistance in grapevine (*Vitis vinifera* L.) parental germplasm under *in-vitro* and natural field conditions

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Abstract

Powdery mildew (PM) caused by *Erysiphe necator* Schwein [(syn. *Uncinula necator* (Schweinf.) Burrill.)] is an ascomycete biotrophic fungus of grapevine, leading to substantial yield and economic losses in infected crop. Thirty-four genotypes of *Vitis vinifera* L., including commercial varieties and hybrids, two genotypes of *V. champini*, three interspecific hybrids of *Vitis* species and two wild indigenous Himalayan species, namely *V. parviflora* and *V. Jacquemontii* were assessed for resistance to powdery mildew using the leaf disc method *in-vitro* with *E. necator* isolate and in the field under natural condition during 2022 and 2023. Significant differences in the scores among the grape genotypes were observed in both *in-vitro* and field conditions. Resistance evaluation under both conditions classified *V. parviflora*, 110R, Couderc 1613, Dogridge, St. George and *V. vinifera* cultivars like Male Hybrid, Pusa Navrang, Blank Prince and Merlot as resistant sources, which can serve as valuable donor parents for breeding programs. Correlation analysis illustrated the negative correlation between disease severity index (DSI) and temperature ($r = -0.50$ (max), $r = -0.48$ (min)), rainfall ($r = -0.07$) and a positive correlation with relative humidity ($r = 0.42$). *In-vitro* inoculation, *Organisation Internationale de la Vigne et du Vin Descriptors* (OIV) scores (7 days post inoculation (dpi)) showed a negative correlation with hyphal area percentage ($r = -0.90$) and a positive correlation with necrosis ($r = 0.86$) indicating the resistant genotypes showing necrosis at the infection site and thus restricted the pathogen growth.

Keywords: Disease severity index (DSI), powdery mildew resistance, genotypes, OIV scoring.

Introduction

The grapevine (*Vitis* spp.) is one of the world's most cultivated horticultural crops all over the world (Otto et al. 2022). It has a great export value and economic potential due to higher yields (Calonnec et al. 2004). The production of grapes is limited due to several pest and disease infestations of which powdery mildew is the second most important endemic disease, causing huge economic losses, i.e., 20-40% reduction in yield every year. At present, its management is largely dependent upon the use of chemical fungicides. Rising production costs generate health and environmental concerns, potentially promoting the selection of *E. necator*-resistant genotypes (Kunova et al. 2021). Traditional phenotyping is based on the evaluation of resistance or susceptibility after natural (field) and artificial inoculation on leaves or leaf discs.

The screening for disease is performed by many researchers at the field intensity to observe the level of incidence in genotypes (Wan et al. 2007; Shikari et al. 2014; Atak et al. 2016; Tetali et al. 2018). The differences

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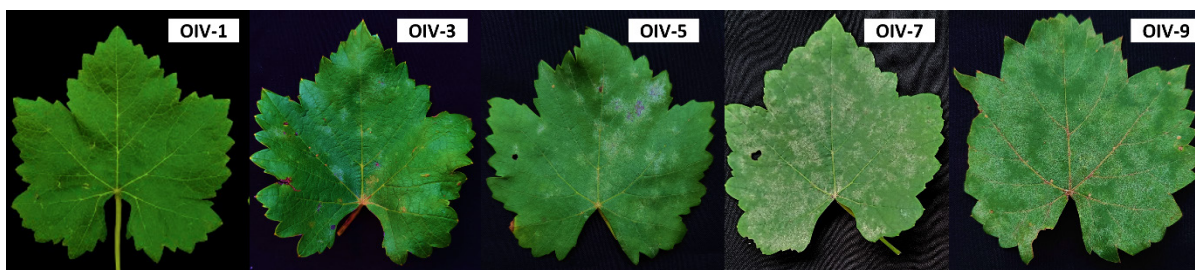
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Table 1. A list of genotypes used in the study

S.No.	Genotype	Species/ parentage	Sl. No.	Genotype	Species/ parentage
1	Anab-E-Shahi	<i>V. vinifera</i>	22	R ₂ P ₁₉	<i>V. vinifera</i> (Pearl-of-Csaba x Beauty Seedless)
2	Alumwick	<i>V. vinifera</i>	23	R ₂ P ₃₆	<i>V. vinifera</i> (Pearl-of-Csaba x Beauty Seedless)
3	Banqui Abyad	<i>V. vinifera</i>	24	Pusa Aditi (H-75-32)	<i>V. vinifera</i> (Banqui Abyad x Per-75-32)
4	Bharat Early	<i>V. vinifera</i>	25	Perlette	<i>V. vinifera</i>
5	Black Muscat	<i>V. vinifera</i>	26	Pusa Navrang	<i>V. vinifera</i> (Madeleine Angevine x Rubi Red)
6	Black Prince	<i>V. vinifera</i>	27	Pearl-of-Csaba	<i>V. vinifera</i>
7	Beauty Seedless	<i>V. vinifera</i>	28	Pusa Purple Seedless	<i>V. vinifera</i> -ER R ₂ P ₃₆ (Pearl-of-Csaba x Beauty Seedless)
8	Cardinal	<i>V. vinifera</i>	29	Pusa Seedless	<i>V. vinifera</i>
9	Cabernet Sauvignon	<i>V. vinifera</i>	30	Pusa Swarnika (H-76-1)	<i>V. vinifera</i> (Hur x Cardinal)
10	Centennial Seedless	<i>V. vinifera</i>	31	Pusa Trishar	<i>V. vinifera</i> (Hur x Bharat Early x Beauty Seedless)
11	Chardonnay	<i>V. vinifera</i>	32	Pusa Urvashi	<i>V. vinifera</i> (Hur x Beauty Seedless)
12	Dog Ridge	<i>V. champini</i>	33	Salt Creek	<i>V. champini</i>
13	Fakhri	<i>V. vinifera</i>	34	St. George	<i>V. rupestris</i>
14	Flame Seedless	<i>V. vinifera</i>	35	Syrah	<i>V. vinifera</i>
15	Hur	<i>V. vinifera</i>	36	Tas-A-Ganesh	<i>V. vinifera</i>
16	Jacquemontii	<i>V. Jacquemontii</i>	37	Tempranillo	<i>V. vinifera</i>
17	Julesky Muscat	<i>V. vinifera</i>	38	<i>Vitis parviflora</i>	<i>V. parviflora</i>
18	MACS Punjab Purple (H-516)	<i>V. vinifera</i> (Catawba x Beauty Seedless)	39	Couderc 1613	<i>V. riparia</i> x <i>V. cinerea</i>
19	MA x BS	<i>V. vinifera</i>	40	1103 Paulsen	<i>V. berlandieri</i> x <i>V. rupestris</i>
20	Merlot	<i>V. vinifera</i>	41	H-70-56	<i>V. vinifera</i> (Hur x Beauty Seedless)
21	Male Hybrid	<i>V. vinifera</i> (Banqui Abyad x Victory)	42	110 Richter	<i>V. champini</i>

**Fig. 1:** Field screening of grape genotypes using *Organisation Internationale de la Vigne et du Vin Descriptors* (OIV) i.e., OIV455-1 descriptors

in susceptibility to powdery mildew are influenced by environmental and cultural factors and phenological stages concerning the intensity of infection. *In-vitro* inoculations overcome some of the drawbacks, although they may not fully reproduce the host-pathogen system that occurs naturally. The inoculation procedures have been employed in the studies of several grapevine diseases and the evaluation of the pathogenicity of several fungal species. *In-vitro* methods that utilize selection agents in controlled

conditions i.e., pathogens, could be useful in breeding programs and may offer the plant breeders an additional tool for identifying disease-resistant plants (Švabova and Lebeda 2005). The laboratory inoculation assay is useful for plant breeders to correlate the results well with the field responses. The *in-vitro* inoculation using leaf disc method was followed in this study to assess the interaction between the pathogen *E. necator* and grape genotypes easily.

The germplasm originated and is native to temperate

Table 2. OIV (OIV 455-1) descriptions for grape powdery mildew

OIV score	Disease symptoms
1	Very low (tiny spots or no symptoms, neither visible sporulation nor mycelium <i>i.e.</i> , 0.1-5.0% of the whole leaf)
3	Low (limited patches < 2 cm in diameter, limited sporulation and mycelium <i>i.e.</i> , 5.1-30.0% of the whole leaf)
5	Medium (patches usually limited with a diameter of 2-5 cm <i>i.e.</i> , 30.1-45.0% of the whole leaf)
7	High (vast patches with limited strong sporulation and abundant mycelium <i>i.e.</i> , 45.1-85.0% of the whole leaf)
9	High (vast patches with limited strong sporulation and abundant mycelium <i>i.e.</i> , 45.1-85.0% of the whole leaf)

zones of North America. Grape breeders investigated it for sources of powdery mildew (PM) resistance (Barker et al. 2005). Powdery mildew resistance has also been explored in *V. rotundifolia* (syn. *M. rotundifolia*), *V. rupestris*, *V. riparia* and *V. aestivalis* to be more resistant to PM than cultivated European *V. vinifera* cultivars (Cadle-Davidson et al. 2011). Developing and deploying novel powdery mildew resistant varieties is considered one of the most promising strategies toward sustainable Viticulture. An investigation was carried out to evaluate the prediction accuracy of the field responses by the *in-vitro* (laboratory) screening assay of grapevines to *E. necator*, to explore the durability of field and *in-vitro* screening for two years and to identify grapevine genotypes resistant to powdery mildew. It is expected that the information generated may be helpful in grapevine breeding in order to achieve eco-friendly resistance to powdery mildew.

Materials and methods

Plant material

Forty two grape genotypes (Table 1) were maintained in the Field Gene Bank at the Division of Fruits and Horticultural Technology, ICAR-IARI, New Delhi, India. The evaluated genotypes consisted of commercial varieties and newly developed hybrids of different *Vitis* species. The vines were maintained following the recommended package of practices. A replication of three vines was taken for the collection of data for two consecutive years *i.e.*, 2022 and 2023.

Disease assessment under natural field conditions

Throughout the two-year investigation, no fungicide applications were done on the experimental plants. The disease severity of all the genotypes grown at the Field Gene Bank was recorded under natural conditions at peak disease incidence (*i.e.* August). Thirty leaves of three vines in

each genotype were observed for the evaluation of natural infection and disease severity index were calculated Each leaf was graded using descriptors recommended by the *Organisation Internationale de la Vigne et du Vin Descriptors* (OIV), *i.e.* OIV455-1 for powdery mildew (1-9 scores) as shown in Fig. 1. and grades were then converted into a disease severity index by using the formula as suggested by Wan et al. (2007). Phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and broad sense heritability (h^2b) were estimated according to Okelola et al. (2007).

The grades were then converted into a DSI by using the formula;

$$DSI = \frac{[\text{Sum of (Grade value x No. of leaves in that grade)}]}{\text{Total leaf No. x No. of leaves in that grade}} \times 100$$

Disease Assessment under Artificial Conditions by using Leaf Disk Method

The leaf discs of each genotype were artificially inoculated (*in-vitro*) with *Erysiphe necator* as per the procedure suggested by Zandler et al. (2021). Three leaf discs per genotype were subjected to surface sterilization with 3% (w/v) sodium hypochlorite solution for 5 minutes. Followed by three rinses in sterile double-distilled autoclaved water and followed by drying on blotting paper. The leaves were then subjected to punching with cork borer and were inoculated with a fungal spore suspension of an *E. necator* isolate (15 mL sterile water with 10 μ L Tween 20, *i.e.*, 1×10^5 /mL) originating from susceptible Cabernet Sauvignon leaves and the petri dishes were sealed with Parafilm®. After inoculation, the shoots were incubated in a growth chamber (20–25°C temp and relative humidity 50–85% RH). Leaf scoring of leaf discs was performed at four days and seven days post inoculation (dpi) assigned by the inverse of OIV scores and the percentage of leaf disc area covered by hyphae (20, 40, 60 and 80%) and necrosis formation was observed under Olympus CX33 microscope under visible

Table 3. Resistance levels of grape genotypes based on Disease Severity Index

DSI value	Disease reaction
0.0 - 0.9	Immune
1.0 - 5.0	Extremely resistant
5.1 - 10.0	Highly resistant
10.1 - 25.0	Resistant
25.1 - 40.0	Moderately resistant
40.1 - 55.0	Moderately susceptible
55.1 - 70.0	Susceptible
70.1 - 85.0	Highly susceptible
85.1 - 100.0	Extremely susceptible

Table 4. Necrosis formation scores

Score	Necrosis
0	No necrosis
1	Random necrosis associated with appressoria formation
2	Necrosis at primary hyphae
3	Necrosis is associated with nearly all appressoria formation

light.

The necrosis formation after inoculation is associated with appressoria formation on the adaxial surface of the leaf and it is scored on a scale of 0 to 3.

Statistical Analysis

Disease severity scores from the laboratory and field evaluations were estimated after using analysis of variance (ANOVA) and Tukey's HSD test applied to detect statistically

Table 5. Disease Severity Index for powdery mildew in grape under subtropical region

Genotype	Disease Severity Index (DSI)			Disease reaction (field screening)	Disease reaction (<i>In-vitro</i> screening)
	2021-22	2022-23	Mean		
Anab-e-Shahi	51.25 ± 3.31 ^e	54.15 ± 1.09 ^e	52.70 ± 2.18 ^d	MS	4.67
Alumwick	81.11 ± 3.15 ^{bc}	83.02 ± 4.18 ^{ab}	82.06 ± 1.03 ^b	HS	1.33
Banqui Abyad	33.89 ± 1.20 ^f	36.63 ± 1.87 ^f	35.26 ± 1.13 ^e	MR	6.67
Bharat Early	35.28 ± 2.10 ^f	38.05 ± 0.90 ^f	36.66 ± 0.62 ^e	MR	6.00
Black Muscat	64.17 ± 4.73 ^d	67.30 ± 5.87 ^d	65.73 ± 4.30 ^c	S	2.50
Black Prince	22.50 ± 2.2 ^h	23.91 ± 3.61 ^{hi}	23.21 ± 1.67 ^f	R	7.50
Beauty Seedless	46.67 ± 1.44 ^e	53.52 ± 5.16 ^e	50.09 ± 3.27 ^d	MS	4.00
Cardinal	37.78 ± 0.87 ^f	38.23 ± 3.4 ^f	38.00 ± 1.28 ^e	MR	6.50
Centennial Seedless	36.94 ± 0.24 ^f	37.91 ± 4.22 ^f	37.43 ± 2.03 ^e	MR	5.67
Cabernet Sauvignon	90.14 ± 1.34 ^a	87.25 ± 1.64 ^{ab}	88.69 ± 1.49 ^a	HS	2.33
Dog Ridge	22.78 ± 1.73 ^h	23.92 ± 2.96 ^{hi}	23.35 ± 0.69 ^f	R	7.67
Chardonnay	45.14 ± 2.55 ^f	46.37 ± 5.21 ^f	45.76 ± 3.43 ^e	MS	6.17
Fakhri	79.86 ± 0.24 ^c	80.67 ± 3.2 ^{bc}	80.26 ± 1.61 ^b	HS	1.67
Flame Seedless	51.11 ± 0.24 ^e	53.64 ± 2.95 ^e	52.38 ± 1.48 ^d	MS	4.67
Hur	77.64 ± 5.55 ^c	78.68 ± 6.66 ^{bc}	78.16 ± 2.52 ^b	HS	1.33
Jacquemontii	33.75 ± 3.15 ^{gh}	35.58 ± 4.11 ^{ghi}	34.66 ± 0.70 ^f	MR	6.17
Julesky Muscat	33.61 ± 5.69 ^f	36.93 ± 1.78 ^f	35.27 ± 3.49 ⁺	MR	6.67
MACS Punjab Purple	33.61 ± 1.27 ^{gh}	34.43 ± 1.76 ^{ghi}	34.02 ± 0.52 ^f	MR	6.50
MA x BS	51.94 ± 1.92 ^e	53.18 ± 2.02 ^e	52.56 ± 0.33 ^d	MS	4.33
Merlot	22.08 ± 1.44 ^h	23.49 ± 2.5 ^{hi}	22.79 ± 1.90 ^f	R	7.50
Male Hybrid	1.87 ± 0.7 ⁱ	3.63 ± 0.38 ^j	2.75 ± 0.43 ^g	ER	8.83
R ₂ P ₁₉	68.19 ± 2.44 ^d	71.15 ± 1.48 ^{cd}	69.67 ± 1.23 ^c	S	2.67
R ₂ P ₃₆	37.22 ± 1.46 ^f	36.53 ± 2.67 ^f	36.88 ± 2.01 ^e	MR	6.00
Pusa Aditi	50.00 ± 3.00 ^e	52.31 ± 4.1 ^e	51.15 ± 3.18 ^d	MS	4.00
Perlette	65.42 ± 1.91 ^d	67.49 ± 2.48 ^d	66.46 ± 0.29 ^c	S	2.67
Pusa Navrang	22.92 ± 1.10 ^h	23.59 ± 1.19 ^{hi}	23.25 ± 0.83 ^f	R	7.67
Pearl-of-Csaba	36.39 ± 2.37 ^f	34.37 ± 3.22 ^{fg}	35.38 ± 0.95 ^e	MR	6.00
Pusa Purple Seedless	35.42 ± 3.00 ^f	36.48 ± 2.38 ^f	35.95 ± 2.22 ^e	MR	6.33
Pusa Seedless	79.44 ± 3.64 ^c	82.40 ± 0.83 ^{ab}	80.92 ± 1.71 ^b	HS	2.67
Pusa Swarnika	33.19 ± 3.59 ^f	35.75 ± 1.22 ^f	34.47 ± 1.60 ^e	MR	6.33
Pusa Trishar	89.03 ± 1.68 ^{ab}	92.08 ± 1.72 ^a	90.56 ± 1.53 ^a	ES	1.67

Pusa Urvashi	51.25 ± 3.7 ^e	53.81 ± 1.57 ^e	52.53 ± 1.76 ^d	MS	4.33
Salt Creek	33.47 ± 4.28 ^f	37.12 ± 1.94 ^f	35.29 ± 2.44 ^e	MR	6.00
St. George	22.08 ± 2.92 ^h	25.03 ± 2.73 ^{ghi}	23.55 ± 2.82 ^f	R	7.50
Syrah	34.58 ± 3.15 ^f	37.65 ± 2.35 ^f	36.12 ± 1.18 ^e	MR	6.00
Tas-A-Ganesh	35.00 ± 1.82 ^f	37.59 ± 1.6 ^f	36.30 ± 1.00 ^e	MR	6.67
Tempranillo	20.69 ± 2.06 ^h	21.91 ± 3.01 ⁱ	21.30 ± 0.75 ^f	R	7.50
<i>Vitis parviflora</i>	2.83 ± 0.82 ⁱ	2.92 ± 0.06 ^j	2.88 ± 0.39 ^g	ER	8.33
Couderc 1613	23.61 ± 1.34 ^{gh}	22.39 ± 2.25 ⁱ	23.00 ± 1.19 ^f	R	7.50
1103 Paulsen	31.81 ± 3.01 ^{fg}	33.34 ± 4.57 ^{gh}	32.58 ± 1.33 ^e	MR	7.00
H-70-56	35.83 ± 1.82 ^f	36.34 ± 2.57 ^f	36.09 ± 0.79 ^e	MR	7.00
110 Richter	7.64 ± 0.64 ⁱ	8.02 ± 0.88 ^j	7.83 ± 0.67 ^g	HR	7.83
C.V. (%)	6.20	7.02	5.24		

Values expressed in mean ± se and the following different letter down the column are significantly different using Tukey's HSD test. ER = Extremely resistant, HR = Highly resistant, R = Resistant, MR = Moderately resistant, S = Susceptible, HS = Highly susceptible and ES = Extremely susceptible significant differences by R Studio software with R.4.3. The relationships between laboratory and field (natural infection) evaluations were analyzed using Spearman's rank correlation (Spearman 1904). Correlations (r) were also computed by using R studio.

Results

Field screening under natural conditions varied significantly ($p < 0.01$) among different grapevine genotypes over the two years of evaluation (2022 and 2023). The disease severity scores ranged from 2.75 (highly resistant) to 88.69 (highly susceptible) in the genotypes studied. When disease severity index scores for different years were compared, it was found that most of the genotypes exhibited consistent disease responses during the two years. Variations in ranking between field and *in-vitro* evaluations were observed.

By using both laboratory and field disease assessments (Table 5), *V. parviflora*, 110R, Male Hybrid showed high resistance to *E. necator* ranging between DSI of 5.1-10.0 and with score OIV-9. The genotypes like Couderc 1613, St. George, Pusa Navrang, Black Prince, Dogridge and Merlot showed resistance ranging between DSI 10.1 to 25.0 and OIV score of 8. The 17 genotypes, namely, Bharat Early, MACS Punjab Purple (H-516), Banqui Abyad, Cardinal, Centennial Seedless, Julesky Muscat, R₂P₃₆, Pearl of Csaba, Pusa Purple Seedless, Pusa Swarnika, Salt Creek, Syrah, Tas-A-Ganesh, 1103 Paulsen, *V. Jacquemontii*, Tempranillo and H-70-56 showed moderate resistance with DSI ranging between 25.1 to 40.0 and OIV score 7. Most of the commercially cultivated cultivars showed different susceptibility ranges for *V. vinifera*, moderately susceptible to extremely susceptible reactions. Anab-e-Shahi, Beauty Seedless, Flame Seedless, MA x BS, Pusa Aditi, Pusa Urvashi and Chardonnay were moderately susceptible with DSI ranging between 40.0-55.0 and OIV score of 5. The DSI between 55.1 to 70.0 and OIV score 3 were classified as susceptible and the genotypes like Black

Muscat, hybrid R₂P₁₉ and Perlette were rated as susceptible to *E. necator*. Cabernet Sauvignon, Hur, Alumwick, Fakhri, Pusa Seedless and Pusa Trishar were showed high susceptibility with high disease severity index ranging from 70.1-85.0 and OIV-1. The relationship between field and laboratory screening indicates a significant correlation with Spearman's rank coefficient of 0.98. These results confirmed that the laboratory leaf disc method is reliable for screening powdery mildew resistance of grapevine cultivars/lines and their hybrids.

Powdery mildew severity in field screening showed a significant positive correlation with relative humidity ($r = 0.42$), while it was negatively correlated with an average minimum temperature ($r = -0.48$) and average maximum temperature ($r = -0.50$) during disease occurrence (Fig. 2). Air temperature of 26 to 29°C and 10 to 13°C, maximum and minimum temperature respectively is found optimum for pathogen development. The maximum incidence of powdery mildew was observed in August and continued to increase (Fig. 2b). The incidence of powdery mildew was recorded low when temperature increased from 30°C and maximum disease incidence was observed at 65 to 70% relative humidity. After *in-vitro* inoculation, the data of hyphal growth, necrosis and OIV scores (4 and 7 dpi) were compared with the help of a correlation plot (Fig. 3). Here, a significant positive correlation was observed for the necrosis formation ($r = 0.86$) at 4 and 7 dpi. In addition, a negative correlation for a percentage of hyphal area coverage on leaf disc ($r = -0.90$) was observed for four and seven dpi. The percentage of hyphal area coverage on leaf disc ($r = -0.80$) showed a negative correlation with necrosis formation in all the parameters observed. The strongest negative correlation was observed at 4 dpi, indicating a small negative effect of necrosis formation on hyphal growth on the leaves.

When disease severity index scores (field screening) and OIV scores (*in-vitro* screening) were used for the estimation

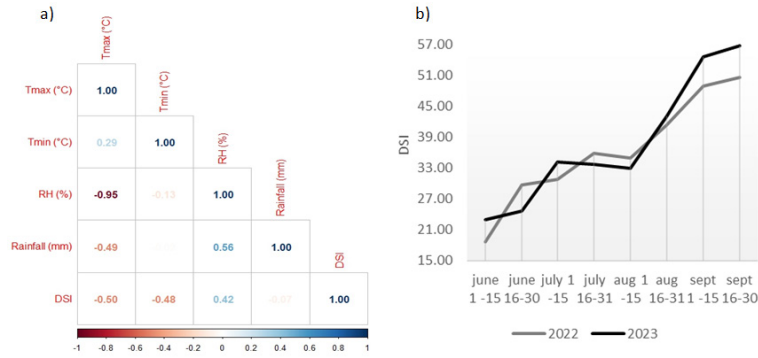


Fig. 2. a) Correlations among weather parameters and Disease Severity Index (DSI) for powdery mildew in grape. b) Disease severity index of powdery mildew during 2022 and 2023

Table 6. Genetic analysis of disease severity under field and in-vitro conditions in 2022 and 2023

Genetic parameter (%)	Field		<i>In-vitro</i>	
	2022	2023	2022	2023
Phenotypic coefficient of variation	54.60	53.10	42.67	38.17
Genotypic coefficient of variation	54.25	52.64	39.63	32.91
Broad sense heritability	0.98	0.98	0.86	0.74

of the phenotypic coefficient of variation (PCV), genotypic coefficient of variation (GCV) and broad sense heritability (h^2) for both the years, the marginal differences were found between PCV and GCV, suggesting a minor role of the environment in expression of powdery mildew resistance (Table 6). The relative high heritability was found in field screening with 0.98 and 0.98, while in *in-vitro* screening, it ranged from 0.86 and 0.74 for both years, respectively for powdery mildew incidence.

Discussion

Only a few resistant and moderately resistant grapevine genotypes with varying resistance to powdery mildew disease have been reported in India. Therefore, the identification of novel sources of resistance to disease has been a major objective for many researchers involved in breeding programs (Fiyaz et al. 2014; Lukšić et al. 2022). The present study observed a significant variation in the degree of disease susceptibilities among 42 grapevine genotypes. Of these, the genotypes like *V. parviflora*, 110R, Male Hybrid, Coudere 1613, St. George, Pusa Navrang, Black Prince, Dogridge and Merlot exhibited resistance based on disease severity index, OIV score, percentage hyphal growth and necrosis. Results presented here revealed that all the genotypes significantly differed in the level of resistance under both natural and artificial inoculation conditions. Results of artificial *in-vitro* inoculation on different grape

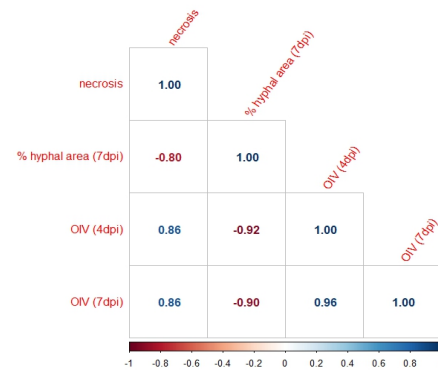


Fig. 3. Correlation plot of the percentages of hyphal area (7 dpi), necrosis formation (4 dpi) and OIV score (4 and 7 dpi). Positive correlations are indicated with blue, while negative correlations are indicated with red. Significance level: $p < 0.05$; all correlations were significant

genotypes were in agreement with those of natural field screening. Similar to the findings recorded in this study, it has been reported that in *V. vinifera* cultivars resistance levels of genotypes vary and generally have low disease resistance. The average inverse *in-vitro* OIV scores suggest that under field conditions of the successive years 2022 and 2023, resistance is determinant against powdery mildew. Various research workers have earlier evaluated grape genotypes for powdery mildew resistance using different assessment criteria. Most of the results obtained in the present study were consistent with previous studies (Gujar et al. 2015; Tetali et al. 2018), but some genotypes exhibited differential responses. Among them, *Vitis parviflora* and 110 Richter have always been found resistant to PM in field conditions (Gurjar et al. 2015).

Temperature is the main environmental factor determining the severity of this disease. Maximum incidence of powdery mildew was observed at 26 to 29°C and 10 to 13°C, maximum and minimum temperature respectively under Delhi conditions. Earlier, Thind et al. (2004) found most suitable temperature is around 20 to 25°C for the development of disease and 20 to 27°C (optimum

24–25°C) is favorable for conidial germination and disease development, although germination can occur between 6 to 33°C. In this study, field screening showed a significant positive correlation with relative humidity, while it was negatively correlated with average minimum and maximum temperature during disease occurrence. The higher disease severity is observed with the temperature range of 25–29°C.

The hyphal growth and multi-septate conidiophores were observed in both resistant and susceptible *Vitis* species. The susceptible genotypes showed widespread sporulation; resistant genotypes had scattered sporulation with low-density conidiophores. However, these findings showed that there is indeed an interaction between the necrosis and hyphal area percentage, showing that necrosis formation leads to some extent of the inhibition of powdery mildew progression. The data for the two traits, *i.e.*, percentage of hyphal area and necrosis formation, were reported to have a negative effect of necrosis formation on hyphal growth (Zendler et al. 2021). In the present study, wild species, *Vitis parviflora* and genotypes 110 Ritcher, Male Hybrid, Couderc 1613, St. George, Pusa Navrang, Black Prince, Dogridge and Merlot were found as resistant source can serve as a useful breeding material for improving the powdery mildew disease resistance.

Author's contribution

Conceptualization of research programme (SKS); Designing of the experiment (SKS, JP, CK, GPM, AM, AK); Contribution of experimental materials required (SKS); Execution of field and lab experiments and data collection (MRS, SKS); Analysis and interpretation of data (MRS, SKS, CK, APK); Preparation of the manuscript (MRS, SKS, JP).

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References

- Barker C. L., Donald T., Pauquet J., Ratnaparkhe M. B., Bouquet A., Adam Blondon A. F., Thomas M. R. and Dry I. 2005. Genetic and physical mapping of the grapevine powdery mildew resistance gene, *Run1*, using a bacterial artificial chromosome library. *Theor. Appl. Genet.*, **111**: 370–377. <https://doi.org/10.1007/s00122-005-2030-8>.
- Cadle-Davidson L., Chicoine D. R. and Consolie N. H. 2011. Variation within and among *Vitis* spp. for foliar resistance to the powdery mildew pathogen *Erysiphe necator*. *Plant Dis.*, **95**: 202–211. <https://doi.org/10.1094/PDIS-02-10-0092>.
- Fiyaz R. A., Krishnan S. G., Rajashekara H., Yadav A. K., Bashyal B. M., Bhowmick P. K., Singh N. K., Prabhu K. V. and Singh A. K. 2014. Development of high throughput screening protocol and identification of novel sources of resistance against bakanae disease in rice (*Oryza sativa* L.). *Indian J. Genet.*, **74**(4): 414–422. <https://doi.org/10.5958/0975-6906.2014.00864.5>.
- Gadoury D. M., Cadle-Davidson L., Wilcox W. F., Dry I. B., Seem R. C. and Milgroom M. G. 2012. Grapevine powdery mildew (*Erysiphe necator*): A fascinating system for the study of the biology, ecology and epidemiology of an obligate biotroph. *Mol. Plant Pathol.*, **13**(1): 1–16. <https://doi.org/10.1111/j.1364-3703.2011.00728.x>.
- Gurjar P. S., Singh S. K., Singh A. K., Verma M. K. and Beniwal S. 2015. Screening and biochemical studies on grape genotypes for powdery mildew infection under sub-tropical conditions. *Ind. J. Agric. Biochem.*, **28**(2): 178–182.
- Kunova A., Pizzatti C., Saracchi M., Pasquali M. and Cortesi P. 2021. Grapevine powdery mildew: Fungicides for its management and advances in molecular detection of markers associated with resistance. *Microorganisms*, **9**(7): 1541. <https://doi.org/10.3390/microorganisms9071541>.
- Ledbetter C. A. and Lee S. A. 2019. Maintaining product quality in raisins throughout the introgression of powdery mildew resistance. *J. Hortic. Sci. Biotechnol.*, **94**(4): 460–467. <https://doi.org/10.1080/14620316.2018.1561213>.
- Lukšić K., Zdunić G., Hančević K., Mihaljević M. Ž., Mucalo A., Maul E., Riaz S. and Pejić I. 2022. Identification of powdery mildew resistance in wild grapevine (*Vitis vinifera* subsp. *sylvestris* Gmel Hegi) from Croatia and Bosnia and Herzegovina. *Sci. Rep.*, **12**(1): 1–13. <https://doi.org/10.1038/s41598-022-06037-6>.
- Okelola F. S., Adebisi M. A., Kehinde O. B. and Ajala, M. O. 2007. Genotypic and phenotypic variability for seed vigour traits and seed yield in West African rice (*Oryza sativa* L.) genotypes. *Am. J. Sci.*, **3**(3):34–41.
- Otto M., Geml J., Hegyi Á. I., Hegyi-Kaló J., Pierneef R., Pogány M., Kun J., Gyenesei A. and Váczy K. Z. 2022. *Botrytis cinerea* expression profile and metabolism differs between noble and grey rot of grapes. *Food Microbiol.*, **106**: 104037. <https://doi.org/10.1016/j.fm.2022.104037>.
- Shikari A. B., Rajashekara, H., Khanna A., Krishnan S. G., Rathour R., Singh U. D., Sharma T. R., Prabhu K. V. and Singh A. K. 2014. Identification and validation of rice blast resistance genes in Indian rice germplasm. *Indian J. Genet.*, **74**(3): 286–299. doi: 10.5958/0975-6906.2014.00846.3
- Spearman C. 1904. The proof and measurement of association between two things. *Am. J. Psychoanal.*, **15**:72–101.
- Vábová L. and Lebeda A. 2005. *In-vitro* selection for improved plant resistance to toxin-producing pathogens. *J. Phytopathol.*, **153**(1): 52–64. <https://doi.org/10.1111/j.1439-0434.2004.00928.x>.
- Teh S. L., Fresnedo-Ramírez J., Clark M. D., Gadoury D. M., Sun Q., Cadle-Davidson L. and Luby J. J. 2017. Genetic dissection of powdery mildew resistance in interspecific half-sib grapevine families using SNP-based maps. *Mol. Breed.*, **37**(1): 1–16. <https://doi.org/10.1007/s11032-016-0586-4>.
- Tetali S., Karkamkar S. P. and Phalake S. V. 2018. Grape breeding for powdery mildew resistance. *Indian J. Hortic.*, **75**(4): 541–545.
- Thind T. S., Arora J. K., Mohan C. and Raj P. 2004. Epidemiology of powdery mildew, downy mildew and anthracnose diseases of grapevine. *Diseases of Fruits and Vegetables Volume I: Diagnosis and Management Book*. pp. 621–638.
- Wan Y., Schwaninger H., He P. and Wang Y. 2007. Comparison of resistance to powdery mildew and downy mildew in Chinese wild grapes. *Vitis - J. Grapevine Res.*, **46**(3): 132–136.
- Zendler D., Töpfer R. and Zyprian E. 2021. Confirmation and fine mapping of the resistance locus *Ren9* from the grapevine cultivar 'Regent.' *Plants*. **10**(1): 1–20. <https://doi.org/10.3390/>