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POWDERY MILDEW INDUCED PHYSIOLOGICAL AND BIOCHEMICAL CHANGES IN PEA (*Pisum sativum* L.)

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Plants respond to external stimuli by altering metabolic pathways, resulting in a cascade of different interrelated changes in physiological and biochemical processes. The incidence of powdery mildew in pea induces change in reducing sugars; non-reducing sugars, total sugars, electrolyte leakage and plant dry weight. Powdery mildew resistant and susceptible pea genotypes respond differentially to these traits. Powdery mildew resistant genotypes had comparatively less electrolyte leakage and less concentration of reducing, non-reducing and total sugars before and after infection. A significant increase in electrolyte leakage, non-reducing and total sugars was induced in susceptible genotypes after powdery mildew infection. The reduction in plant dry weight and reducing sugars after infection was more pronounced in powdery mildew susceptible and resistant genotypes respectively. All the traits being significantly correlated with disease severity can be an important component of an effective powdery mildew screening procedure in pea.

Keywords: Electrolyte leakage, legumes, metabolism, reducing sugars, screening

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INTRODUCTION

Plants are continuously exposed to different diseases caused by a range of pathogens. The incidence of plant diseases is the major limiting factor for sustainable crop yields (Agrios, 1988; Azmat *et al*., 2010). Plant-microbe interaction results in the alteration of different physiological and biochemical attributes of plants. The changes in physiological and biochemical attributes collectively affect the magnitude and direction of metabolism and reduce the photosynthesis efficiencies (Swarbrick *et al*., 2006; Jobic, 2007).

Pea (*Pisum sativum*) is a rich source of plant protein for human food and animal feed (Cousin, 1997; Azmat, 2011). The incidence of powdery mildew (*Erysiphe pisi*) causes severe decline in the grain yield of pea. It is believed that the reduction in yield and deterioration of pea seed quality is due to certain changes in different physio-biochemical traits (Gritton and Ebert, 1975; Jones and Dangl, 2006; Swarbrick *et al*., 2006). The differential yield response of powdery mildew resistant and susceptible pea plants under the prevalence of disease (Gritton and Ebert, 1975) suggests that they may have the same response to different physio-biochemical traits. There exists a negative correlation between grain yield and powdery mildew resistance in pea (Munjal *et al*., 1963). Usuallay field screening is done for the selection of powdery mildew resistant pea plants and it is difficult to distinguish between the escape and resistance through field screening (Azmat *et al.,* 2012a). Therefore, it would be helpful to develop more robust methods and/to validate the results of the field screening with other affordable techniques. Since the development of powdery mildew resistant pea genotypes is a lengthy process, therefore, the selection of resistant plant(s) with reliability is a prerequisite for success.

The objective of the current research was to check the effect of powdery mildew on different physio-biochemical traits in resistant and susceptible pea genotypes. The possibility of these traits *viz.,* electrolyte leakage, reducing sugars, non-reducing sugars, total sugars and plant dry weight as selection criteria for powdery mildew resistance was also assessed.

MATERIALS AND METHODS

Plant material and powdery mildew inoculation: Seed of 30 pea genotypes (Table 1) was surface sterilized with 2% hypochlorite and rinsed thoroughly with sterile water. The seeds were planted in 10 × 10 cm plastic pots containing thoroughly washed and sterilized vermiculite. Two seeds per pot were used for all the genotypes. Approximately 200 mL of nutrient solution containing 1 mM $CaCl_2.2H_20$, 100 μM KCl, 800 μM $MgSO_4.7H_2O$, 10 μM Fe EDTA, 35 μM H_3BO_3, 9 μM $MnCl_2.4H_2O$, 0.8 μM $ZnCl_2$, 0.5 μM $Na_2MoO_4.2H_2O$, 0.3 μM $CuSO_4.5H_2O$, 800 μM KH_2PO_4 , 700 μM Na_2HPO_4 and 1 mM NH_4NO_3 was added to each pot (Allaway *et al*., 2000; Azmat *et al*., 2013). Ten pots for each genotype were maintained; five pots were inoculated while five were kept as un-inoculated. The pea genotypes used were from the same maturity group (Azmat *et al.,* 2011). The pots were placed in growth chamber at 22°C

Table 1. The percentage of fluctuations of different physio-biochemical traits in response to powdery mildew among pea genotypes.

Sr. No.	Genotype	% Δ RS	% Δ NRS	% Δ TS	% Δ EL (48 HAI)	% Δ EL (72 HAI)	% Δ EL (96 HAI)	% Δ EL (120 HAI)	% Δ DW (30DAI)
1	**9057***	**-20.0**	**69.7**	**15.1**	**3.4**	**-2.8**	**-0.4**	**3.9**	**-1.93**
2	**9370**	**-19.0**	**71.8**	**15.8**	**11.5**	**0.2**	**0.7**	**-0.6**	**-6.61**
3	**9375**	**-20.6**	**73.7**	**6.9**	**1.2**	**2.0**	**3.0**	**0.2**	**-0.58**
4	**10609**	**-22.1**	**69.6**	**5.4**	**0.8**	**1.0**	**-1.7**	**1.8**	**2.83**
5	**10612**	**-22.1**	**72.7**	**10.1**	**6.5**	**7.1**	**-3.6**	**12.0**	**-0.90**
6	10649$^{\wedge}$	-6.0	172.9	39.5	114.6	110.6	134.8	349.2	-67.38
7	18293	-7.2	141.1	31.4	106.9	125.0	125.9	283.4	-69.81
8	**18412**	**-22.9**	**75.7**	**14.1**	**5.3**	**2.2**	**9.2**	**8.0**	**-3.89**
9	**19598**	**-23.9**	**76.9**	**14.4**	**0.7**	**1.5**	**-2.8**	**6.1**	**3.33**
10	**19611**	**-24.3**	**71.0**	**12.3**	**4.8**	**-0.1**	**0.3**	**1.6**	**-0.17**
11	19616	-7.2	189.9	47.9	113.9	87.2	129.6	272.8	-58.24
12	19727	-7.8	142.9	33.7	119.3	115.0	92.7	144.5	-18.80
13	19750	-9.5	146.8	33.6	81.6	124.1	136.1	156.2	-23.37
14	**19782**	**-25.1**	**70.8**	**5.5**	**-0.5**	**6.7**	**-0.3**	**5.1**	**-2.05**
15	20126	-8.1	206.9	48.5	93.0	105.4	174.8	282.9	-59.46
16	**20152**	**-25.1**	**80.9**	**9.1**	**1.7**	**6.9**	**5.1**	**5.5**	**0.70**
17	**20171**	**-24.9**	**72.7**	**10.1**	**1.1**	**2.2**	**5.8**	**0.2**	**-0.55**
18	9800-10	-6.1	236.9	59.7	117.9	110.6	203.0	366.4	-81.59
19	9800-5	-6.0	258.0	63.6	127.5	117.8	182.8	410.7	-82.88
20	Climax	-6.3	266.9	68.2	168.3	110.9	180.3	430.2	-86.19
21	**It-96**	**-25.0**	**70.7**	**2.3**	**-13.6**	**2.0**	**-0.3**	**-0.4**	**0.79**
22	KQP-6121	-2.6	214.9	57.9	83.4	110.1	116.8	355.2	-73.85
23	KQP-6173	-6.0	236.9	63.0	125.7	122.5	126.0	361.1	-76.51
24	KQP-6185	-6.4	262.9	69.9	144.4	117.4	163.9	388.0	-79.59
25	Meteor-VRI	-6.5	206.8	52.0	154.3	149.7	203.4	425.9	-80.84
26	**No. 267**	**-24.5**	**70.9**	**4.5**	**-0.7**	**2.3**	**1.9**	**0.8**	**-0.75**
27	**No. 380**	**-22.1**	**75.9**	**10.5**	**4.6**	**-0.4**	**-0.1**	**1.1**	**1.16**
28	P1	-6.8	212.8	56.6	116.6	124.9	199.2	458.8	-66.97
29	PF-400	-7.3	270.0	69.0	196.8	151.8	173.1	439.7	-86.97
30	Premium	-4.9	266.0	73.6	123.8	125.6	171.5	413.5	-74.66
Average	Combined	-14.2	148.5	33.5	67.2	64.6	84.3	186.1	-36.5
	Resistant	**-23.0**	**73.1**	**9.7**	**1.9**	**2.2**	**1.2**	**3.2**	**-5.0**
	Susceptible	-6.6	214.5	54.3	124.3	119.3	157.1	346.2	-67.9

* =The values for resistant genotypes are given in bold font, ^=The values for susceptible genotypes are given in normal font. Sign "-"represent decrease in value. The abbreviations used are % Δ= Percent change, RS= Reducing sugars, NRS= Non-reducing sugars, TS=Total sugars, EL= Electrolyte leakage, DW=Plant dry weight, HAI= Hours after infection, DAI= Days after infection

with a 14:10 h light: dark photoperiod with light intensity of 400 µmol m^{-2} s^{-1}.

All the genotypes were inoculated with powdery mildew at 8th node stage. The powdery mildew inoculum was homogenized in tween-20 and water-agar solution (Reeser *et al.*, 1983; Azmat *et al.,* 2012b) containing approximately 4×10^4 conidia·mL^{-1}. After inoculation, the pots were placed in the chamber with controlled environment that provided a 14 h light period at 22°C and a 10 h dark period at 15°C. The relative humidity was maintained at 60% and the photon flux density was 400 µmol m^{-2} s^{-1}. Control without inoculation was kept in the separate chamber with the similar specifications to avoid cross contamination.

***Disease severity*:** A 1-5 scale based on percentage of leaf area affected with disease was used to classify pea genotypes as resistant and susceptible to powdery mildew. The data on disease severity were recorded 15 DAI (days after inoculation). The scale based on percentage of leaf area affected with powdery mildew is given as under: 1 = 0-5% infection (Resistant), 2 = 6-20% infection (Moderately resistant), 3 = 21-60% infection (Moderately susceptible), 4 =61–80% infection (Susceptible), 5 = 81–100% infection (Highly susceptible). The cultivar Meteor-VRI was used as susceptible check, its infection response was "highly susceptible" with a disease score of 5.

***Electrolyte leakage assay*:** Electrolyte leakage assay was employed to assess the extent of membrane damage in powdery mildew affected pea leaves. Five discs (1 cm) were cut from the middle part of powdery mildew inoculated and non-inoculated leaves from five plants of each genotype 48, 72, 96 and 120 HAI (Hours after Inoculation). The leaf discs were floated on distilled water (30 ml) in Falcon tubes overnight (Ádám *et al.,* 2000). The Changes in conductivity of the bathing solutions were measured with an EC meter (TOA-CM-14P, Japan).

***Estimation of sugars*:** The extract of uninoculated and powdery mildew affected fresh leaves in 80% ethanol (Jaypal and Mahadevan, 1968) was used for estimating reducing, non-reducing and total sugars in pea 15 DAI. Nelson's modification of Somogyi's method (Nelson, 1944) was used for estimating reducing sugars. Total sugars were estimated by hydrolyzing non-reducing sugars using 1N H_2SO_4 and then quantified as in case of reducing sugars (Nelson, 1944). Non-reducing sugars were calculated by subtracting the reducing sugars from that of total sugars.

***Measurement of plant dry weight*:** The data on plant dry weight (g) was recorded from powdery mildew inoculated and non-inoculated plants of all the genotypes 30 DAI. The individual plants were placed in oven at 70°C till constant weight then the data were recorded accordingly.

***Statistical analysis*:** Three independent biological experiments were conducted for each trait; all the values presented here are the averages. The data recorded for each experiment had significant homogeneity and hence it was pooled for statistical analysis. The changes in the response to powdery mildew by resistant and susceptible genotypes with reference to control were quantified in terms of percentage for all traits. The percentages were calculated as increase and decrease of the parameter under consideration by using following formula:

$$\%\,\Delta = \frac{(\mathit{Value\ of\ control\ sample} - \mathit{Value\ of\ inoculated\ sample})}{\mathit{Value\ of\ control\ sample}} \times 100$$

Pearson correlation between traits was performed for control and infected genotypes. A UPGMA (Unweighted Paired Group Method with Arithmetic Mean) based phenogram was constructed with Euclidian distance. Statistical analyses were carried out using SPSS 17 (SPSS, Chicago, I L), MVSP 3.1 (Kovach Computing Services, Anglesey, Wales), and Microsoft Excel (QI Macros).

RESULTS

All the traits, reducing sugars, non-reducing sugars, total sugars, electrolyte leakage and dry weight have shown highly significant (p>0.01) variation among all the genotypes (Table 2).

The 30 pea genotypes were classified into five main disease response groups based on disease score ranging from 1-5

Table 2. Measures of dispersion and central tendency of different physio-biochemical traits under powdery mildew infection and control condition.

Trait		Min.		Max.		Ave.		SD		CV		SE	
		R*	S$^\wedge$	R	S	R	S	R	S	R	S	R	S
RS (mg/g)	Control	5.1	13.31	6.3	15.9	5.6	14.7	0.4	0.7	0.1	0.0	0.1	0.2
	Infected	3.9	12.27	5.1	14.9	4.3	13.7	0.3	0.8	0.1	0.1	0.1	0.2
NRS	Control	2.1	5.06	3.9	6.0	2.9	5.5	0.6	0.3	0.2	0.1	0.2	0.1
(mg/g)	Infected	3.6	12.29	6.8	21.8	5.0	17.5	1.0	3.4	0.2	0.2	0.3	0.8
TS	Control	7.2	18.37	10.3	21.5	8.5	20.2	0.9	0.9	0.1	0.0	0.2	0.2
(mg/g)	Infected	7.5	24.56	11.9	36.0	9.3	31.3	1.3	3.9	0.1	0.1	0.3	1.0
EL(µS)	Control	10.1	15.4	16.2	19.3	12.9	16.7	2.0	1.3	0.2	0.1	0.5	0.3
(48 HAI)	Infected	10.5	30.6	16.1	47.3	13.1	37.3	1.8	4.9	0.1	0.1	0.5	1.2
EL(µS)	Control	14.4	19.8	23.6	24.9	17.6	22.7	2.8	1.5	0.2	0.1	0.8	0.4
(72 HAI)	Infected	14.0	41.6	24.1	57.2	18.0	49.7	3.0	4.6	0.2	0.1	0.8	1.2
EL(µS)	Control	17.8	29.5	29.5	39.8	21.6	35.1	4.1	2.9	0.2	0.1	1.1	0.7
(96 HAI)	Infected	17.8	72.2	29.4	109.4	21.9	90.3	4.1	13.9	0.2	0.2	1.1	3.5
EL(µS)	Control	22.1	38.4	41.7	45.1	27.4	42.8	6.4	2.0	0.2	0.0	1.7	0.5
(120 HAI)	Infected	22.2	103.5	42.1	243.5	28.2	191.9	6.1	45.6	0.2	0.2	1.6	11.4
DW (g)	Control	1.1	1.8	3.3	3.7	2.4	2.9	0.5	0.5	0.2	0.2	0.1	0.1
(30DAI)	Infected	1.0	0.5	3.2	2.2	2.3	0.9	0.5	0.5	0.2	0.6	0.1	0.1
PM	Control	*There was no infection*											
Infection %	Infected	0.0	28.0	17.0	96.0	6.4	73.4	5.1	23.2	0.8	0.3	1.4	5.8

* =The values for resistant genotypes are given in bold font, ^=The values for susceptible genotypes are given in normal font. The abbreviations used are RS= Reducing sugars, NRS= Non-reducing sugars, TS=Total sugars, EL= Electrolyte leakage, DW=Plant dry weight, PM= Powdery mildew, Max.= Maximum, Min= minimum, Ave=Average, SD= Standard deviation, CV= Coefficient of variability, SE= Standard error, R=Resistant, S= Susceptible.

(Fig. 1). Six genotypes (10609, 10612, 19782, 20171, It-96 and No.267) were highly resistant having “1” disease score. Nine genotypes (9800-10, 9800-5, Climax, KQP-6121, KQP-6185, Meteor-VRI, P1, PF-400 and Premium) were placed in highly susceptible group with disease score “5”. There were eight genotypes in “moderately resistant” group, five in “moderately susceptible” and two in “susceptible” group, respectively (Fig. 1).

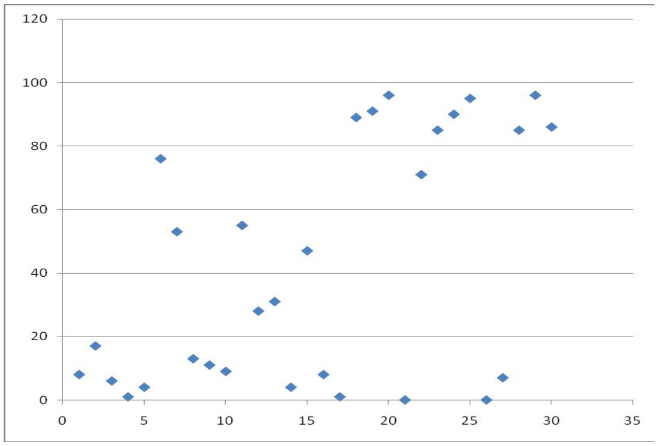

Figure 1. Scatter plot showing the differential response of 30 pea genotypes to powdery mildew 15 DAI. The number of genotypes corresponds to the serial numbers given in Table 1, respectively.

Significant variation was observed among all genotypes for electrolyte leakage under infected and control conditions at 48, 72, 96 and 120 HAI (Table 2). The susceptible genotypes had more leakage of electrolytes than resistant genotypes before and after powdery mildew infection. Under un-inoculated conditions, powdery mildew susceptible pea genotypes had 34-68% more leakage of electrolyte than resistant. The comparison of electrolyte leakage under infection with control have shown an average increase of 67.2%, 64.6%, 84.3% and 186.1%, respectively, for 48, 72, 96 and 120 HAI for all the genotypes regardless of their disease response (Table 1). The increase in electrolyte leakage was not significant ($p<0.05$) for resistant genotypes. The susceptible pea genotypes have shown highly significant increase in electrolyte leakage ranging from 119.3-346.2% with an average increase of 186.7% (Table 1). Maximum increase in leakage of electrolytes was observed at 120 HAI in both powdery mildew susceptible and resistant genotypes. All the disease response groups behaved differently from each other. Resistant and moderately resistant groups differed non-significantly from each other but were significantly different from other three susceptible groups (Fig. 2a).

There was significant variability among the genotypes for reducing, non-reducing and total sugars 15 DAI both for control and infected conditions. Total sugars ranged from 7.5-36.1 mg.g^{-1} under un-inoculated condition. Reducing and non-reducing sugars ranged from 5.1-15.9 mg.g^{-1} and 2.1-6 mg.g^{-1}, respectively, for control (Table 2). Under disease free conditions susceptible genotypes had 2.63 times more reducing sugars and 1.89 times more non-reducing sugars than resistant genotypes, respectively. Under diseased conditions reducing and non-reducing were 3.18 and 3.5 times more respectively in susceptible than resistant genotypes (Table 2).

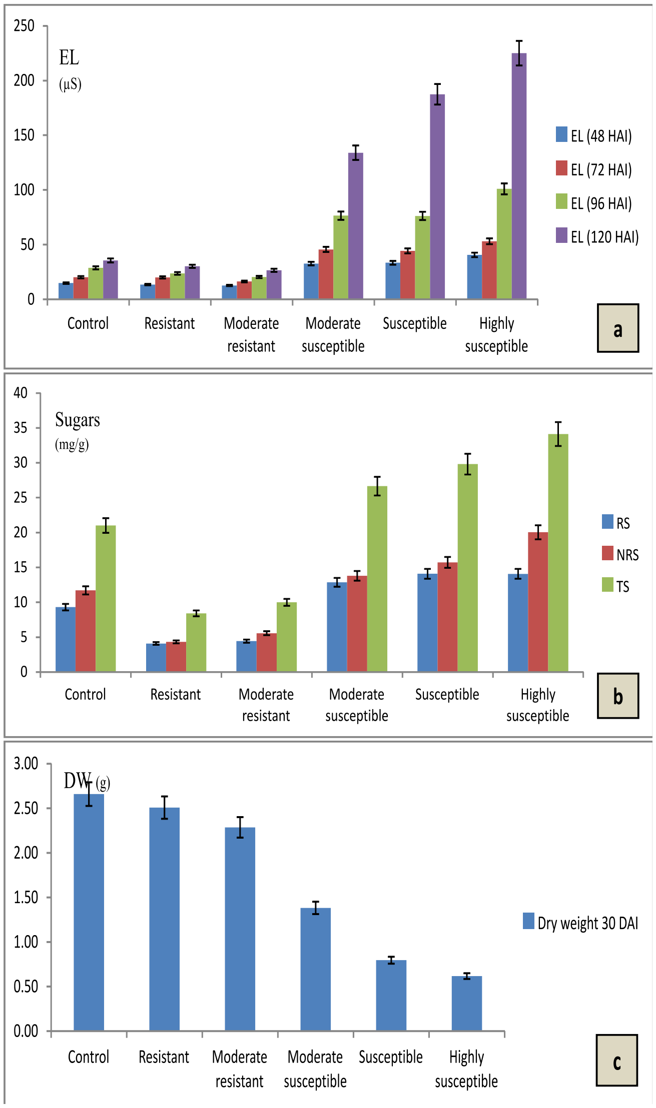

Figure 2. The comparative response of five disease groups of pea genotypes. (a) Electrolyte leakage (EL) 24, 48, 72 and 120 Hours After Infection (HAI); (b) Concentration of reducing (RS), non-reducing (NRS) and total sugars (TS); (c) Plant dry weight (DW) 30 Days After Infection (DAI) in comparison with control after powdery mildew infection.

After powdery mildew infection reduction in reducing sugars ranged from 2.6-25.1%. The average reduction in the concentration of reducing sugars in response to infection was more in resistant genotypes (23%) than susceptible genotypes (6.6%) (Table 1). An increase in non-reducing sugars was observed in all genotypes 15 DAI ranging from 69.6-270%. Avergae increase in non-reducing sugars was less in resistant genotypes (73.1%). The increase in non-reducing sugars was significantly higher in susceptible genotypes ranging from 141-270%. (Table 1). The average concentrations of reducing, non-reducing and total sugars were significantly different ($p>0.05$) among the disease groups. Both resistant groups were non-significantly different ($p<0.05$) from each other and were significantly different from three susceptible groups (Fig. 2b).

The change in plant dry weight was recorded for all genotypes 30 DAI with reference to control. Under control conditions, plant dry weight ranged from 1.1-2.9 g, which was non-significantly different among powdery mildew resistant and susceptible genotypes. Significant reduction ranging from 18.8-87% was recorded in plant dry weight for susceptible genotypes 30 DAI. Non-significant reduction in dry weight was recorded for resistant genotypes with an average reduction of 5% (Table 1). Powdery mildew resistant and susceptible groups can be easily differentiated on the basis of plant dry weight (Fig. 2c).

The Pearson correlation analysis indicated that all the traits have highly significant positive correlation with disease severity. UPGMA and Euclidian distance based phenogram constructed using all the traits, classified 30 genotypes in two main clusters. The cluster-I comprised of all the powdery mildew resistant genotypes while clusters-II had

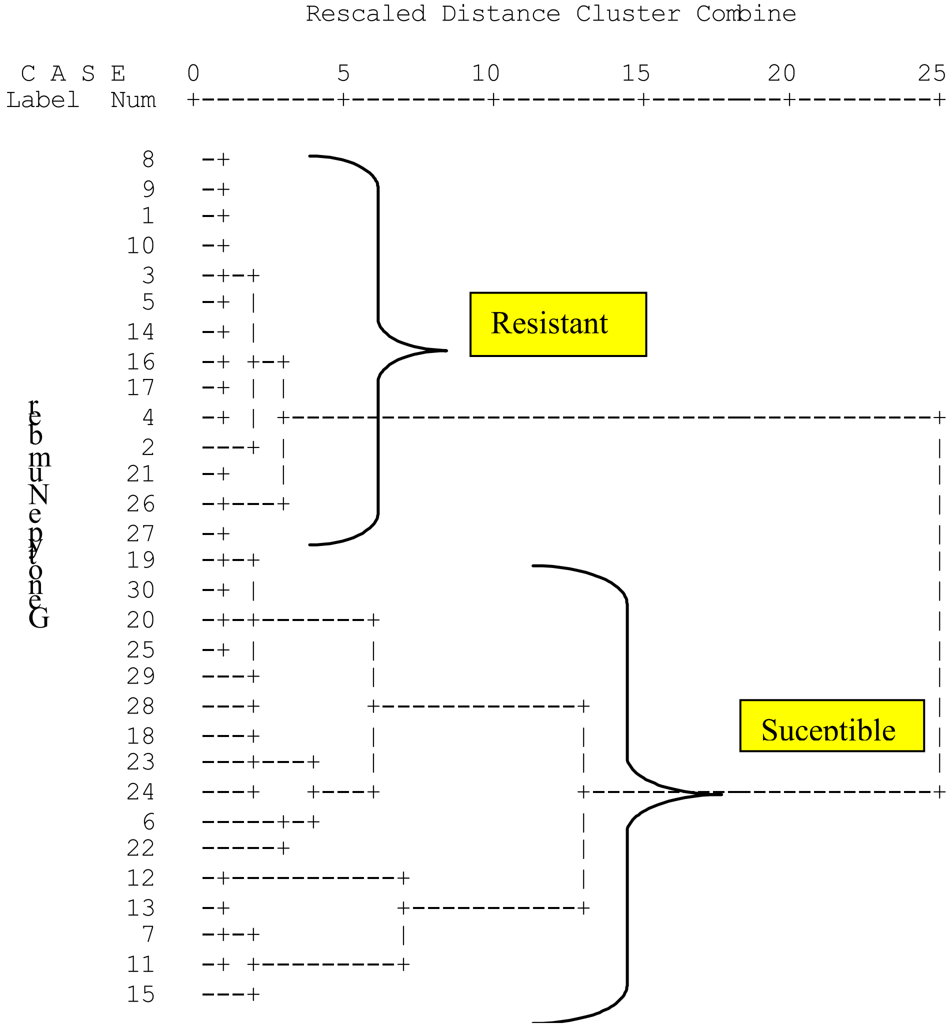

Figure 3. Euclidean distances and physio-biochemical traits based UPGMA-Phenogram showing independent grouping of resistant and susceptible pea genotypes. The number of genotypes corresponds to the serial numbers given in Table 1, respectively.

Table 2. Measures of dispersion and central tendency of different physio-biochemical traits under powdery mildew infection and control condition.

Trait		Min.		Max.		Ave.		SD		CV		SE	
		R*	S^	R	S	R	S	R	S	R	S	R	S
RS (mg/g)	Control	5.1	13.3	6.3	15.9	5.6	14.7	0.4	0.7	0.1	0.0	0.1	0.2
	Infected	3.9	12.3	5.1	14.9	4.3	13.7	0.3	0.8	0.1	0.1	0.1	0.2
NRS (mg/g)	Control	2.1	5.1	3.9	6.0	2.9	5.5	0.6	0.3	0.2	0.1	0.2	0.1
	Infected	3.6	12.3	6.8	21.8	5.0	17.5	1.0	3.4	0.2	0.2	0.3	0.8
TS (mg/g)	Control	7.2	18.4	10.3	21.5	8.5	20.2	0.9	0.9	0.1	0.0	0.2	0.2
	Infected	7.5	24.6	11.9	36.0	9.3	31.3	1.3	3.9	0.1	0.1	0.3	1.0
EL(µS) (48 HAI)	Control	10.1	15.4	16.2	19.3	12.9	16.7	2.0	1.3	0.2	0.1	0.5	0.3
	Infected	10.5	30.6	16.1	47.3	13.1	37.3	1.8	4.9	0.1	0.1	0.5	1.2
EL(µS) (72 HAI)	Control	14.4	19.8	23.6	24.9	17.6	22.7	2.8	1.5	0.2	0.1	0.8	0.4
	Infected	14.0	41.6	24.1	57.2	18.0	49.7	3.0	4.6	0.2	0.1	0.8	1.2
EL(µS) (96 HAI)	Control	17.8	29.5	29.5	39.8	21.6	35.1	4.1	2.9	0.2	0.1	1.1	0.7
	Infected	17.8	72.2	29.4	109.4	21.9	90.3	4.1	13.9	0.2	0.2	1.1	3.5
EL(µS) (120 HAI)	Control	22.1	38.4	41.7	45.1	27.4	42.8	6.4	2.0	0.2	0.0	1.7	0.5
	Infected	22.2	103.5	42.1	243.5	28.2	191.9	6.1	45.6	0.2	0.2	1.6	11.4
DW (g) (30DAI)	Control	1.1	1.8	3.3	3.7	2.4	2.9	0.5	0.5	0.2	0.2	0.1	0.1
	Infected	1.0	0.5	3.2	2.2	2.3	0.9	0.5	0.5	0.2	0.6	0.1	0.1
PM Infection %	Control	*There was no infection*											
	Infected	0.0	28	17.0	96	6.4	73.4	5.1	23.2	0.8	0.3	1.4	5.8

* =The values for resistant genotypes are given in bold font, ^=The values for susceptible genotypes are given in normal font. The abbreviations used are RS= Reducing sugars, NRS= Non-reducing sugars, TS=Total sugars, EL= Electrolyte leakage, DW=Plant dry weight, PM= Powdery mildew, Max.= Maximum, Min= minimum, Ave=Average, SD= Standard deviation, CV= Coefficient of variability, SE= Standard error, R=Resistant, S= Susceptible.

susceptible genotypes (Fig. 3). Both the clusters have significantly different values for the physio-biochemical traits focused in this manuscript (Table 2).

DISCUSSION

Erysiphe pisi the causal organism of pea powdery mildew is an obligate biotroph. Powdery mildew induced plant defense mechanisms and the alteration in primary metabolism leads to changes in secondary metabolism (Bonfig *et al.*, 2006). The fluctuation in primary metabolites causes reduction in yield and deteriorates the quality of harvest. The changes in chlorophyll concentrations in response to pea powdery mildew have already been reported by the authors (Azmat *et al.*, 2012c). Powdery mildew induces down regulation in photosynthesis (reduction in chlorophyll) and increase the demand for assimilates simultaneously hence affecting the direction and magnitude of metabolism (Pego *et al.*, 2000)

Sugars are not only essential for respiration, growth and bio-synthesis of different compounds, but these can also regulate the expression of photosynthesis related genes (Koch, 1996; Pego *et al.*, 2000; Berger *et al.*, 2004). Powdery mildew infection induces changes in photosynthesis and respiratory pathways causing significant fluctuation in the concentrations and ratios of reducing and non-reducing sugars (Klement and Goodman, 1967). In powdery mildew resistant genotypes the concentration of total, reducing and non-reducing sugars were significantly less than susceptible genotypes (Table 2 & Fig. 2b). A significant change in the concentration of sugars with reference to control was observed in susceptible genotypes after infection. In susceptible genotypes, there was an average increase in non-reducing sugars (214%) and total sugars (54%). There was comparatively more reduction in reducing sugars (23%) in powdery mildew resistant genotypes (Table 1).

Electrolyte leakage is the measure of senescence in response to powdery mildew infection (Heitefuss, 2001). Electrolyte leakage was significantly enhanced in powdery mildew infected leaves of susceptible genotypes while resistant genotypes showed non-significant increase (Table 1). Electrolyte leakage had significant correlation with disease severity. Significant leakage of electrolytes was induced 48 HAI in susceptible genotypes while there was comparatively less electrolyte leakage in resistant genotypes even 120 HAI (Table 2). The observation on electrolyte leakage after powdery mildew infection in susceptible genotypes suggested two possibilities: a) the incidence of disease totally damages the cell membrane or b) powdery mildew infection alters the membrane permeability while the cell membrane remained intact. The differential response of powdery mildew resistant and susceptible genotypes to electrolytes leakage contained in cells (Table 2 & Fig. 2a) should have some definite role in determining the disease response of plants. The electrolytes might have influence on signaling mechanism involved in disease response mechanisms. The incidence of powdery mildew adversely affects the biomass and yield in pea (Gritton and Ebert, 1975). The reduction in biomass was more pronounced in susceptible genotypes (Table 1).

Powdery mildew resistant and susceptible genotypes have shown differential response to the physio-biochemical traits focused in the current research (Table 2 & Figure 3). All the traits have also shown significant correlation with diseases severity, suggesting the possibility of using these traits as reliable criterion for effective screening for powdery mildew in pea.

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