



Research Article

Assessment of Genetic Variability in Onion against Purple Leaf Blotch Disease under Field Conditions and its Management

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Abstract | Purple blotch disease is exerting a severe threat on onion production. The current study was conducted for screening of accessions against purple blotch disease and determining the source of resistance against Purple blotch disease of onion caused by *Alternaria porri*. Potential of 50 onion accessions was evaluated against *A. porri* during 2017 and 2018 under field conditions. After that effect of some fungicides and plant extracts was determined for disease suppression on a susceptible variety of onion. During both years of study, eight accessions (Phulkara, Texas Early, Cylon, Sunset, Red Gystal, Rubi F1, Red Flame, ON-14133) exhibited resistant response with minimum percent disease index (PDI) which ranged between 5-10%, followed by Mirpurkhas, Nasarpuri, HON-1069, Pania, Rubi F2, GSL-132, Red Moon, PK-1032, F-1122, Red Imposta, Early Red, Desi Large and Vrio-4 which showed moderately resistant response to *A. porri* with PDI of 11-20%, whereas, the four accession including Golden Arab, Desired, Sultan F1 and Red Nasik expressed Moderately resistant response with PDI of 21-40%. Moreover, six accessions (Yellow Gystal, Vrio-9, Vrio-5, Vrio-4, Vrio-1, and Vrio-3) expressed susceptible response with high value of PDI 41-60% whereas; nine accessions (CBS-130, ON-14121, Pink Panther, Vrio-7, Vrio-3, F1 Zeus, SE-16, Vrio-8 and Marvi) showed maximum value (Above 60%) of PDI and expressed highly susceptible response to *A. porri*. Among fungicides and botanical extracts, Chlorostrobin and *Azadirachta indica* extract expressed significant reduction in disease severity. It was concluded that out of 50, only eight accessions were resistant which might be grown successfully for higher crop production. Moreover, use of Chlostrobin and extract of *Azadirachta indica* may be used for disease management.

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Keywords | *Allium cepa*, *Alternaria porri*, Accessions, Percent disease Index (PDI), Purple blotch



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Introduction

Onion (*Allium cepa* L.) belongs to family Alliaceae and is cultivated all around the world for its nutritive, medicinal and health protective value. It is mainly known as “Queen of Kitchen” as it accompanies all cooking items (Mandloi, 2017). Onion carries specific taste and numerous medicinal characteristics likewise antifungal, germicide, antispasmodic and antibacterial (Griffiths *et al.*, 2002). China and India are the leading onion producing countries (Mallor *et al.*, 2011). In Pakistan about 147.2 hectare is under onion cultivation with 1,981.7 thousand tons production (PES, 2017-18). Onion leaves are also utilized as supplementary nourish for cattle, rabbits and poultry birds (Mba and Akueshi, 2001). Onion is attacked by numerous fungal diseases but purple blotch disease caused by *Alternaria porri* is the potential extortion to the successful cultivation of onion and is the potential threat all over the world (Kumar and Palakshapra, 2008).

Production of host specific and non-specific toxins significantly influences the level of disease incidence and these toxins cause leaf necrosis in susceptible varieties (Mamgain *et al.*, 2013). It also destroys bulb initiation stimulus and delays bulb maturation. *A. porri* mainly affects bulbs and leaves of the plants which leads to heavy yield losses of up to 97% (Ravichandran *et al.*, 2017; Kareem *et al.*, 2012). Pathogen is soil borne and it remains viable in soil for longer period and causes severe losses to onion crop (Yaradua, 2003). Purple blotch disease is commonly initiated by moderate temperature 25-30°C and high humidity 80-90%. Numerous strategies, including the application of fungicides and plant extracts have been tested against *A. porri* (Jhala *et al.*, 2017a; Yadav *et al.*, 2017; Younas *et al.*, 2021). Among all strategies, some are not economically applicable for the farmers because of their high cost whereas others have direct or indirect deteriorating and degrading effect on human beings and environment (Iglesias *et al.*, 2021). Perfect method to overcome the maladies is the use of resistant onion genotypes. Therefore screening from available germplasm is pre-requisite to determine the accessions which are resistant against *A. porri* (Thaxton and Zik, 2001). Thus, present research was aimed to find out the resistant accessions for the management of purple blotch disease under field conditions and the other component of current study was consisted on the evaluation of suitable fungicides and plant extracts for

disease management in susceptible accessions.

Materials and Methods

Assessment of onion accessions against Alternaria porri under natural field conditions

In first component of the study, Onion nursery was grown at Vegetable Research Area, Ayub Agriculture Research Institute Faisalabad (AARI) on 25th and 21st of October during 2017 and 2018 respectively, whereas the transplanting was done after 45 days of sowing during both years of study. Disease free seeds of fifty accessions were used for nursery raising. The nursery was transplanted in field by following P×P and R×R distance of 22 and 60cm respectively. The experiment was laid out in Randomized complete block design (RCBD) and replicated thrice. Disease intensity were recorded after two months of transplantation by using 0-5 rating scale (Sharma, 1986) and all the accessions were grouped in different ranks depending on their resistance and susceptibility to *A. porri* (Pathak *et al.*, 1986). The percent disease index (PDI) was determined by following formula, proposed by (Wheeler, 1969).

$$PDI = \frac{\text{Total number of numerical ratings}}{\text{Number of observation}} \times \frac{100}{\text{Maximum disease rating}}$$

Assessment of fungicides and botanical extracts against A. porri

In second component of the study, susceptible variety of onion (Marvi) was cultivated to assess the capability of fungicides and botanical extracts against *A. porri* by following the same planting geometry, nursery raising and transplanting dates during both study years as in first component. The same experimental design and number of replications were followed.

Management of purple blotch through fungicides

Two fungicides Chlorostrobin and Nanok at the concentration of 1.5g/ liter of water were evaluated against purple blotch disease of onion under field conditions. Experiment was laid out in randomized complete block design (RCBD) by adopting standard row to row and plant to plant spacing. Three sprats at the interval of fifteen days were used and the data regarding disease reduction was after seven and fourteen days of each spray. First spray application was done after the appearance of characteristics symptoms.

Management of purple blotch through plant extracts

Three plant extracts (*Azadirachta indica*, *Ocimum*

tenuiflorum, *Allium sativum*) were evaluated under natural field conditions. Three sprays of plants extracts were done whereas; the first spray was done after the appearance of initial disease symptom in onion field however, remaining sequential sprays was done at the interval of fifteen days. For the preparation of botanical extracts, 75g fresh leaves were crushed in 25 ml sterilized distilled water using a sterilized mortar and pestle. Resultant standard solution was filtered through sterilized four layered muslin cloth and whatman filter paper No.14, moreover standard arbitrarily is received (Ilyas *et al.*, 1996). Similarly standard concentration was prepared by mixing 100 ml of standard concentration with 100 ml of sterilized water.

Hand sprayer (IHT-401) was used for the application of chemical fungicides and botanical extracts. After the application of chemical fungicides and botanical extracts against *A. porri* disease reduction was calculated by using following formula.

$$\text{Red in DS (\%)} = \frac{\text{DS in control plants} - \text{DS in treated plants}}{\text{DS in contro plants}} \times 100$$

Red= Reduction, DS= Disease Severity.

Statistical analysis

The recorded data from each experiment were statistically analyzed through Fisher's analysis of variance (ANOVA) technique by using Statistix 8.1 software. The treatments means were compared by using Least Significant Difference (LSD) test at 5% probability level.

Table 1: Disease data was noted by visual observation and rating scale as described by (Sharma, 1986).

Disease ratings	Descriptions	Re-sponse
0	No symptoms	I
1	Few spots on tip with 10% covered area	R
2	Purplish brown patches with covering of 20% leaf area	MR
3	Patches with paler outer region covering 40% leaf area	MS
4	Up to 75% leaf area covered with leaf streaks	S
5	Complete leaves are dried	HS

I: Immune/ highly resistant; R: Resistant; MR: Moderately Resistant; MS: Moderately Susceptible; S: Susceptible; HS: Highly Susceptible.

Results and Discussion

Assessment of onion accessions against *Alternaria porri* under natural field conditions

The current study was conducted for two experimental

years during 2017 and 2018 under natural epiphytotic conditions. Assessment and evaluation of fifty onion accessions was carried out against purple leaf blotch disease and results are presented in Table 2. Disease severity (DS) ranged from 5% to79% among onion accessions. Different degrees of resistance were recorded among fifty accessions (Tables 2 and 3). The results revealed that there was not a single accession that showed immune response to *Alternaria porri* during both experimental years (2017-2018). However, during 2017 eight accessions were found resistant and thirteen accessions showed moderately resistant response. Moreover, eleven accessions were moderately susceptible whereas, nine accessions expressed susceptible response. Furthermore, remaining nine screened accessions demonstrated highly susceptible reaction to *A. porri* with percent disease index above 60%. Minimum disease severity was recorded in Phulkara (4.28%) whereas the maximum disease severity was observed in Marvi (79.00%).

Table 2: Response of onion accessions to purple blotch disease under field conditions during (2017).

Sr.	Accessions	PDI (%)	Re-sponse	Sr.	Accessions	PDI (%)	Re-sponse
1	Phulkara	4.28l	R	26	Desi Black	26.31v	MS
2	Texas Early	4.85l	R	27	Robina	26.31v	MS
3	Cylon	5.87k	R	28	Husri	28.12u	MS
4	Sunset	6.30k	R	29	S-4466	32.43t	MS
5	Red gystal	7.14j	R	30	Pussa Red	33.90s	MS
6	Rubi F1	8.10i	R	31	Vrio-06	35.99r	MS
7	Red Flame	8.85hi	R	32	Vrio-10	38.14q	MS
8	ON-14133	9.09h	R	33	SV-748NP	41.35p	S
9	Mirpurkhas	10.98g	MR	34	Perma	42.74o	S
10	Nasarpuri	11.66fg	MR	35	FSD Red	45.32n	S
11	HON-1069	11.85ef	MR	36	Yellow gystal	48.31m	S
12	Pania	12.59e	MR	37	Vrio-09	51.75l	S
13	RubiF2	13.73d	MR	38	Vrio-05	54.93k	S
14	GSL-132	14.82c	MR	39	Vrio-04	55.49k	S
15	Red Moon	15.88b	MR	40	Vrio-1	56.66j	S
16	PK-1032	16.260b	MR	41	Vrio-3	58.78i	S
17	F-1122	16.266b	MR	42	CBS-130	62.77h	HS
18	Red Imposta	17.83a	MR	43	ON-14121	65.51g	HS
19	Early Red	18.06a	MR	44	Pink Panther	67.54f	HS
20	Desi Large	18.90z	MR	45	Vrio-07	69.62e	HS
21	Vrio-4	19.71y	MR	46	Vrio-03	70.61d	HS
22	Golden Arab	22.24x	MS	47	F1 Zeus	71.41d	HS
23	Desired	23.86w	MS	48	SE-16	72.58c	HS
24	Sultan F1	24.07w	MS	49	Vrio-08	75.66b	HS
25	Red Nasik	25.89v	MS	50	Marvi	77.72a	HS
LSD(0.05)		0.79					
SE		0.40					

*Mean values in a column sharing similar letters do not differ significantly as determined by the LSD test ($P \leq 0.05$).

Table 3: Response of onion accessions to purple blotch disease under field conditions during (2018-19).

Sr.	Accessions	PDI (%)	Re-sponse	Sr.	Accessions	PDI (%)	Re-sponse
1	Phulkara	5.30E	R	26	Red Nasik	27.50QR	MS
2	Texas Early	5.60E	R	27	Husri	27.57QR	MS
3	Cylon	6.95D	R	28	Robina	27.92Q	MS
4	Sunset	7.31CD	R	29	S-4466	33.32P	MS
5	Red Gystal	8.20BC	R	30	Pussa Red	35.17O	MS
6	Rubi F1	9.08AB	R	31	Vrio-6	36.26N	MS
7	Red Flame	9.19AB	R	32	Vrio-10	39.49M	MS
8	ON-14133	9.42A	R	33	SV-789NP	42.26L	S
9	Mirpurkhas	11.27Z	MR	34	Fsd Red	43.93K	S
10	HON-1069	12.69Y	MR	35	Perma	44.92K	S
11	Nasarpuri	12.71Y	MR	36	Yellow Gystal	49.48J	S
12	Pania	13.29Y	MR	37	Vrio-9	54.48I	S
13	Rubi F2	15.73X	MR	38	Vrio-4	55.04I	S
14	GSL-132	15.96X	MR	39	Vrio-5	55.29I	S
15	Red Moon	17.09W	MR	40	Vrio-1	58.94H	S
16	PK-1032	17.32W	MR	41	Vrio-3	59.87H	S
17	F-1122	17.70W	MR	42	CBS-130	64.85G	HS
18	Early Red	19.21V	MR	43	ON-14121	65.59G	HS
19	Vrio-4	19.28V	MR	44	Pink Panther	68.27F	HS
20	Red Im-posta	19.50V	MR	45	Vrio-8	70.70E	HS
21	Desi Large	19.89V	MR	46	Vrio-3	72.65D	HS
22	Golden arab	21.07U	MS	47	F1 Zeus	73.18D	HS
23	Sultan F1	25.21T	MS	48	SE-16	74.91C	HS
24	Desired	25.92ST	MS	49	Vrio-8	77.80B	HS
25	Desi Black	26.67RS	MS	50	Marvi	79.67A	HS
LSD (0.05)		1.060					
SE		0.53					

*Mean values in a column sharing similar letters do not differ significantly as determined by the LSD test ($P \leq 0.05$).

The result of the experimental year (2018) revealed that all accessions maintained their status and exhibited consistency in their response (Resistant, Moderately Resistance, Moderately Susceptible, Susceptible and Highly Susceptible) to *A. porri* however, an increased percent disease index in all tested accessions was recorded in comparison to the experimental year 2017 (Table 3).

Management of purple blotch disease of onion through fungicides and plant extracts

The results of second component of study revealed that chemical fungicide Chlorostrobin caused maximum disease inhibition up to 83%, followed by

Nanok (77.40%), *Azadirachta indica* (68%), *Ocimum tenuiflorum* (65%) and *Allium sativum* (61.33%) (Table 4).

Table 4: Assessment of botanical extracts and fungicides against *Alternaria porri* under field conditions.

Sr.	Treatment	Active ingredients	Disease reduction (%)
1	Chlorostrobin	Azoxystrobin + Chlorothalonil	83.00a
2	Nanok	Azoxystrobin + Flutriafol	77.40b
3	<i>Azadirachta indica</i>	Azadirachtin, Azadirachtin	68.00 c
4	<i>Ocimum tenuiflorum</i>	Apigenin, Polyphenols, Anthocyanins and luteolin	65.00d
5	<i>Allium sativum</i>	Alicin	61.33e
6	Control	Water	0.00f
LSD (0.05)		1.12	
SE		0.48	

Purple leaf blotch is the most disparaging disease of onion which inflicts severe damage to aerial parts of the plant (Bal *et al.*, 2019) and lowers the yield potential to great extent (up to 50%) (Jhala *et al.*, 2017b). Disease development depends on the presence of susceptible host, favorable environmental conditions (temperature, rainfall, relative humidity and wind speed) and virulence strain of the pathogen. Under such scenario the economical and long lasting way to save crop from *Alternaria porri* is the use of resistant genotypes. Although the best possible and foremost probable solution is the development of resistant cultivars by inclusion of resistant genes, but it is time taking journey. Therefore, screening of the available accessions is the short term and easy way to identify resistant source. Keeping in view the above facts, in present study fifty accessions of onion were examined against *A. porri* under natural field conditions for two consecutive years. Results of the contemporary study revealed that there was not a single accession that showed immune/ highly resistant response towards onset of disease. Among all accessions Phulkara exhibited resistant response whereas the Marvi showed highly susceptible response. Results of present study are similar as the findings of Mansha *et al.* (2019) that found nonetheless of the varieties immune to disease whereas, the tested cultivar Phulkara displayed resistant response to *A. porri*. Results of our study are also supported by the previous studies of Behera *et al.* (2013) and Ulhaq *et al.* (2014) that there is not a single genotype which

have resistance towards *A. porri*. In contemporary studies, Chlorostrobin expressed maximum disease reduction as it contains chlorothalonil and azoxystrobin which inhibits multi sites of different enzymes. Nanok is an effective fungicide against Basidiomycetes, Oomycetes, Ascomycetes and Deuteromycetes, moreover, it has curative and protectant characteristics and is highly systemic leading to the long term efficacy. It inhibits mycelial growth, respiration, spore germination and maintains normal leaf area which leads to maximum average yield potential (Younas *et al.*, 2021). *Azadirachta indica* has a complex of numerous constituents likewise, nimbin, nimbolide, nimbidin and limonoids and such ingredients play a pivotal role in disease management by the modulation of different genetic pathways and other activities. β -sitosterol and Quercetin were the first polyphenolic flavonoids purified from neem leaves and were known to have antifungal activities (Govindachari *et al.*, 1998). Furthermore, studies revealed that antimicrobial role of neem aqueous extract significantly inhibits the growth of seed borne fungi i.e., *Rhizopus* and *Aspergillus* and sporulating fungi including *C. lunata*, *C. gloeosporioides* and *H. pennisetii* (Mondali *et al.*, 2009) and the results of study revealed that aqueous extract of *Azadirachta indica* showed growth inhibition against *Alternaria porri*. Results are also in line with the findings of Younas *et al.* (2021) that Chlorostrobin exhibits maximum reduction in disease severity. Findings of current study are also in line with Islam *et al.* (2020) who studied the efficacy of different plant extracts against *A. porri*.

Conclusions and Recommendations

From the above findings of screening of accessions and evaluation of fungicides and plant extracts it was concluded that the eight cultivars which were resistant to this disease can be grown for better crop production. Moreover, Chlorostrobin and extract of *Azadirachta indica* can be used for disease suppression in susceptible accessions.

Novelty Statement

Identification of resistant accessions may be helpful in further studies and breeding programs. Moreover, recommended fungicide and botanical extract may also help farmers for timely management of purple leaf blotch disease and higher crop production.

Author's Contribution

Muhammad Younas: Collection of the data and manuscript writing.

Khalid Hussain: Conceived idea, Literature review.

Abdul Ghaffar: Analyzed and compiled the data.

Muhammad Atiq: Provide Resources.

Niaz Hussain: Supervised the whole research.

Muhammad Azeem Khan: Data analysis.

Wasim abbas: Designed research methodology.

Muhammad Nadeem: Helped in data Collection.

Muhammad Irshad: Data Interpretation.

Nasir Ahmad Khan: Edited the manuscript

Muhammad Zubair: Literature review

Conflict of interest

The authors have declared no conflict of interest.

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