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Epidemiological Study of Onion Against Purple Blotch

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Abstract The field experiment was carried out to understand the diseases development of purple leaf bloch of onion with respect to weather conditions during the year 2016 starting from 14th standard week to 22nd standard week at horticultural research station, haveli farm, Bagalkot dt., Karnataka, India. The three different parameters are recorded viz. percent disease index, apparent rate of infection, area under disease progress curve at each standard week. Experiment conducted for purple blotch of onion was dose by artificial spraying of inoculum of pathogen in open field condition to induction of diseases in plants. The results of this study revealed that the PDI value ranging from 0 to 73.33, Apparent infection rate at seven days intervals showed the range between -0.0000303 to 0 was observed and AUDPC value is recorded 2052.195 and it given the initiative that the delay in the sowing increases the severity of onion purple blotch, reduces seed test weight and yield of crop.

Keywords Onion, Purple blotch, Percent disease index, Apparent rate of infection, Area under disease progress curve.

Introduction

Onion (*Allium cepa* L.) belonging to Alliaceae family is one of the most important commercial vegetable crops grown and consummed widely as a spice or a condiment across the world. Onion is supposed to have its origin in the Middle East Asian Countries and introduced in India from Palestine. It belongs to family *Alliaceae* and genus *Allium* with about 300 species. Onions are good source of minerals, vitamins and carbohydrates. It is valued for its distinct pungent flavor and is an essential ingredient for the cooking in many regions. India is one of the major onion producing and exporting countries. Globally, India ranks first in area (1.05 m ha) and second in production (19.4 m t) with a productivity of 16 t/ha [1]. Though India stands second in onion production in the world, its productivity is below the world's av-

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Table 1. 0–9 point rating scale based on leaf area covered by the pustules Mayee and Datar [5].

Scale	Description of the symptoms
0	Absolutely free from infection
1	Small sized lesions on the leaf covering <1% area
2	Small sized lesions on the leaf covering <2-5% area
3	Small sized lesions on the leaf covering <6-10% area
4	Small sized lesions on the leaf covering <12-15% area
5	Small sized lesions on the leaf covering <16-25% area
6	26–40% area covering
7	41–60% area covering
8	61–75% area covering
9	>75% area covered with spot, most of the leaves dired.

Table 2. Percent disease index (PDI) and AUDPC of onion variety against purple blotch of onion. AUDPC[a]: Area under the disease progress curve.

Standard week	Varieties	Percent disease index (PDI)	Reaction	Apparent rate of infection 'r'
14	Arka Kalyan	0.00	R	0
15	Arka Kalyan	5.56	MR	-0.02
16	Arka Kalyan	18.89	MR	-0.00434
17	Arka Kalyan	42.22	MS	-0.0011
18	Arka Kalyan	62.22	S	0.000259
19	Arka Kalyan	70.00	S	-0.0000323
20	Arka Kalyan	71.11	S	-0.0000313
21	Arka Kalyan	72.22	S	-0.0000303
22	Arka Kalyan	73.33	S	–
	AUDPC[a]	2652.195		

erage (19.1 t/ha) due to several production constraints including diseases.

Onion crop is attacked by 66 diseases, of which 10 bacterial, 38 fungal, 6 nematodes, three viral, one phytoplasmal, one phanerogamic plant parasite and seven miscellaneous diseases and disorder. The major bacterial diseases are: Bacterial flower stalk and leaf necrosis (*Pantoea agglomerans*) fungal diseases are: Purple blotch (*Alternaria porri*) and stemphylium leaf blight (*Stemphylium vesicarium*), viral diseases are: Yellow dwarf (yellow dwarf virus) and nematode diseases are: Stem and bulb nematode (*Ditylenchus dipsaci*) and root knot nematode (*Meloidogyne incognita*). Among these diseases, the purple blotch (*A. porri*) is one of the major constrains in onion cultivation. It causes hige losses in different parts of India. The severity of disease is much more where crop is predisposed by thrips injury Mishra et al. [2]. The pathogen is polyphagus infecting crop like onion, garlic, shallot and other *Allium* crops. High relative humidity (80 to 90%) and potimum temperature (24+2°C) are needeed. In favorable conditions, epidemic may cause total faillur of the crop. Severe losses were recorded due to purple blotch disease Shahanaz et al. [3]. Mishra et al. [2] and that in seed crop has been reported to the extent of 97% Lokra [4].

The diseased plant is characterized by typical symptoms of the disease appears on foliage and on foliage sheath small white sunken spots develop on the leaves which enlarge, become zonate and under moist conditions, turn purple and are also prominent on the inflorescence and stalks. Infection can cause a semi watery rot on necks of bulbs that turn yellow red in color. Infected bulb tissues eventually become papery. Present field experiment was undertaken to know the apparent rate of infesction r in onion cultivar Arka Kalyan from date of sowing to harvest i.e. is 14^{th} standard week to 22^{nd} standard week and also studied on percent disease index, area under the disease progress curve to know the effect of weather condition in disease progress.

Materials and Methods

A field experiment was carried out to assess the effect of sowing dates on purple bltch of onion appearance and development. The trail was carried in plot design with a pot size of 1×0.75 M and plant spacing of 30 × 15 cm. Popular varieties viz., Arka Kalyan cultivar were growth in Haveli, Bagalkot, Karnataka. General agronomical practices were followed for as for the package of practise. Recommended doses of NPK were applied @ 150:50:80 kg/ha. Vermicompost was applied at the time of field preparation @ 20 q/ha. Hand weeding was carried out in all the plots. Observations were recorded at first appearance of the disease symptoms on leaves till the harvest at weekly intervals. Screening was done on 0–9 point rating scale based on leaf

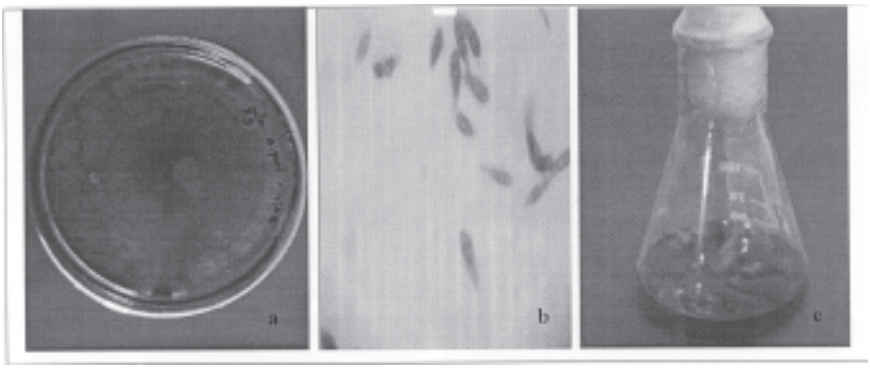

Fig. 1. The growth of the *Alternaria pori* in PDA, in b microscopic view of the pathogen and in c growth of *Alternaria pori* in PDB.

area covered by the pustules [5].

The experiment conducted for purple blotch of onion was done by artificial spraying of inoculum of pathogen in open field condition to induction of diseases in plants.

Isolation and identification

Infested onion leaves showing typical symptoms of purple blotch of onion were collected from the fields, brought to the laboratory and washed thoroughly with distilled water. These leaves were then blot dried and cut with sharp sterilized blade into small bits (5 mm), keeping half healthy and half-diseased portion intact. These pieces were surfaces sterilized with 0.1% aqueous solution of mercuric chloride ($HgCl_2$) for two minutes and then washed by giving three changes with sterile distilled water to remove the traces of mercuric chloride and blot dried. The surface sterilized diseased leaf bits were then inoculated on the solidified and cooled PDA (potato dextrose agar) medium in petri plates under aseptic conditions. Inoculated plates were then incubated in BOD incubator at $24 + 2^{o}C$ temperature. After seven to eight days of incubation, the well-developed mycelial growth, free from any contaminant was obtained. Following hyphal-tip technique, the fungus was transformed/subcultured aseptically on the PDA slants in culture tubes. Through frequent sub-culturing, the fungus was purified and pure culture was maintained given in Figure 1.

Pathogenicity test

Seeds of onion cultivar Arka Kalyan, were surface sterilized with 0.1% HgC12 and sown @ 10 seeds/pot in the earthen pots (25 cm dia.) filled with steam sterilized potting mixture of soil: sand: FYM (2:1:1). Five healthy growing onion seedlings per pot were maintained, watered regularly and kept in the green house for further development. The test pathogen (*A. porri*) was multiplied on the basal culture medium PDA in petri dishes. Spore-cum mycelia suspension of the test pathogen was prepared from 7–8 days old culture in plates by flooding with 5–10 ml sterile distilled water. The resultant spore-cum-mycelial suspension was filtered through double-layered muslin cloth and filtrate obtained was suitably diluted with sterile distilled water to get inoculum's concentration of 5×10^4 spores/ml. Forty days old seedlings of onion cultivar Arka Kalyan already grown in earthen pots were artificially inoculated by spraying with hand sprayer the spore-cum-mycelial suspension (5×10^4 spores/ml) of the test pathogen. Onion seedlings in earthen pots sprayed with sterile water (without inoculums) were maintained as uninoculated suitable control. Pots (both inoculated and uninoculated) were incubated

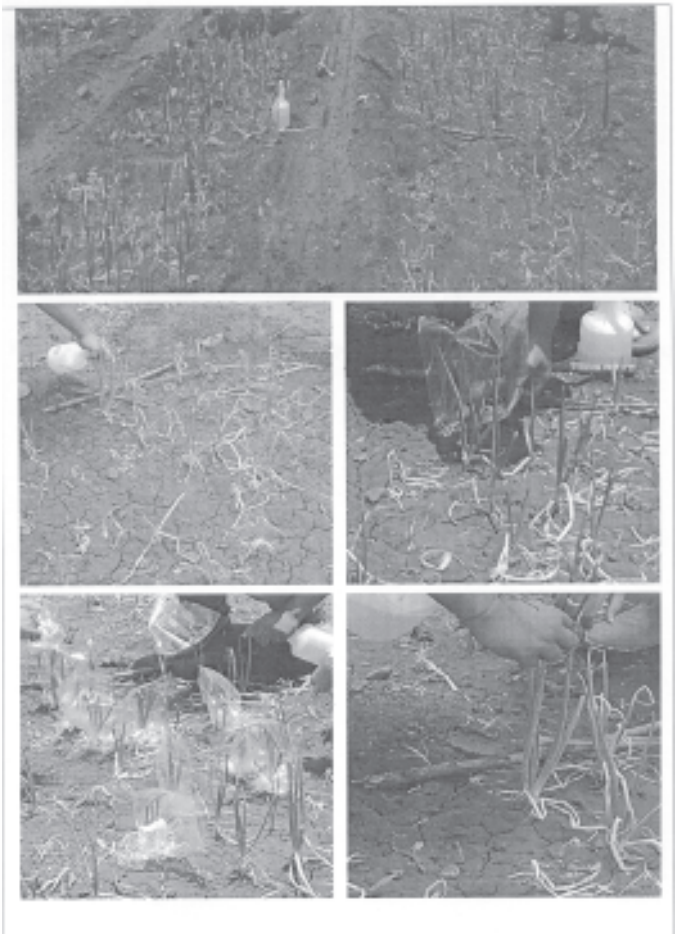

Fig. 2. The field wear experiment was conducted along with the pathogen inoculation and bagging of inoculated with plastic bag.

in the screen house, where high humidity (80–90%) and optimum temperature (24+2°C) were maintained for further development of purple blotch symptoms.

Reisolation

The pathogen was reisolated on basal medium (PDA) from the culture medium artificially inoculated onion seedling showing typical symptoms of purple blotch. Growth of the fungus obtained was transferred on potato dextrose agar slants and compared with original pure culture of the test fungus obtained from naturally infected leaf of onion.

Identification of pathogen

Pure culture of the fungus obtained was inoculated aseptically on autoclaved PDA in petri plates and plates were incubated at 24±1°C for a week. On the basis of pathogenicity test, cultural characteristics and microscopy, the test pathogen was identified *A. porri*.

Percent disease index

Purple blotch of onion percent disease index was recorded at seveen days interval starting form first ap-

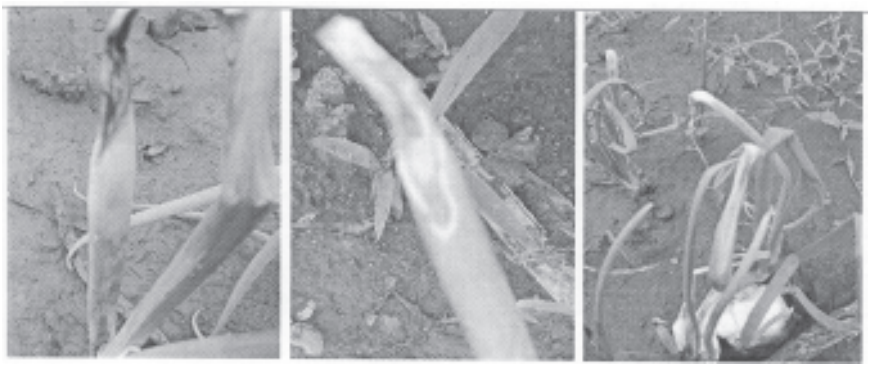

Fig. 3. The diseased symptoms of the onion against purple blotch pathogen.

pearance of the disease by observing the ten of infected plants. Ten plants at bulb developmental stage were randomly selected for scoring the disease at weekly intervals. The severity of purple blotch of onion was recorded by using 0–9 scale [5] given in Table 1.

The screened plants were categorized for their reaction on the basis of PDI values. Those with 10% DI vale were considered as resistant, while those with 11–25% as moderately resistant, 26–50% as moderately susceptible, 51–75 as susceptible and more than 75% as highly susceptible.

Later per cent disease index (PDI) was calculated by using formula given by [6].

$$PDI = \frac{\text{Sum of individual disease rating}}{\text{Total No. of plants observed} \times \text{Maximum disease rating}} \times 100$$

Apparent rate of infection r

The apparent rate of infection (r) at different intervals was calculated by using the formula given by Van der Plank [7] r value is calculated using the equation 1.

$$r = \frac{1}{t_2 - t_1} \log_e \frac{X_2 (1-X_1)}{X_1 (1-X_2)}$$

Where, t is time interval, x is the value of PDI and the $\log_e$ is written as Inverse (IN)

Area under the disease progress curve (AUDPC)

The area under the disease progress curve (AUDPC) was calculated for rating at 14, 15, 16, 17, 18, 19, 20, 21 and 22 standard weeks after inoculation using equation given by Shanner and Finney [8].

$$AUDPC = \sum_{i}^{n-1} (y_i + y_i + 1/2) (t_i+1 - t_i)$$

where y_i is the percent disease index (PDI) observed for the i^{th} treatment, *ti* is the date of the observation and observation were made on *n* dates.

The most commonly used method for estimating the AUDPC, the trapezoidal method, is to discredited the time variable (days) and calculate the average disease intensity between each pair of adjacent time points [9]. We can consider the sample time points in a sequence (*ti*), where the time interval between two time points may be consistent or may vary, and we also have associated measures of the disease level (*yi*).

Results and Discussion

Observations revealed that during the year 2015-16,

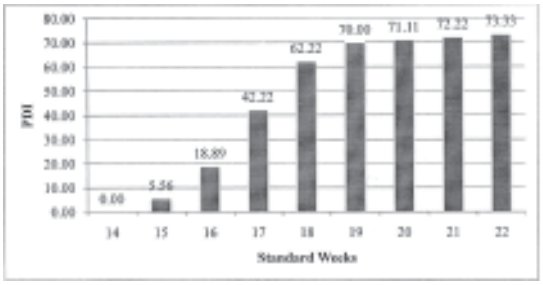

Fig. 4. The per cent disease index (PDI) of onion purple leaf blotch at different time intervals.

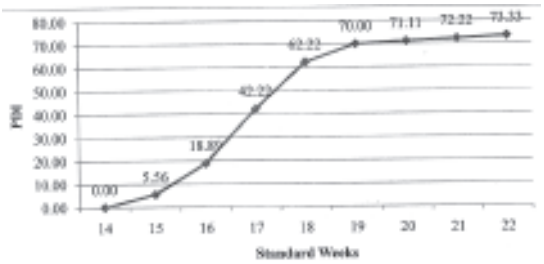

Fig. 5. The per cent disease index (PDI) of onion purple leaf blotch at different time intervals.

the per cent disease index (PDI) of onion purple leaf blotch were recorded, which ranged from 0.00 to 73.33 from the every seven days intervals from 14th standard week to 22nd standard week for the cultivar Arka Kalyan (Table 2, Fig. 2, 3).

In onion the percent disease index was recorded from 14th standard week to 22nd standard week with a seven days intervals. The results of the purple leaf blotch per cent disease index is recorded in first week i.e. is in 14th standard week is 0.00, in second week observed 5.56, in third week reported the 18.89 and from fourth to ninth week recorded the PDI of 42.22, 62.22, 70.00, 71.11, 72.22 and 73.33 respectively. These PDI observed which indicates the increase in disease with favorable condition increases toward the 22th week. With the reaction on the basis of PDI is R, MR, MS, S, S, S, S and S respectively are recorded using 09 point rating scale based on leaf area covered by the pustules given in Table 2, Figure 4, 5. Raju and Naik [10] Chethana et al. [11] and Gupta and Gupta [12] conducted the study in onion and obtained similar results.

The apparent infection rate is an estimate of diseases, based on proportional measures of extent of infection at different times, firstly, a proportional measure of the extent of infection is chosen as the diseases extent metric. Apparent infection r at seven days intervals showed the range between -0.0000303 to 0 was observed. The r value is recorded in first week i.e. is in 14th standard week is 0, in second week observed -0.02, in third week reported the -0.00434, and from fourth to ninth week recorded the apparent infection rate -0.0011, -0.000259, -0.0000323, -0.0000313 and -0.0000303 respectively give in Table 2. In the present investigation the average apparent rate of infection value varied. Similar observation was recorded by Nagesha and Nargund [13] to measure the slow rusting in sunflower.

The area under the disease progress curve (AUDPC) is a useful quantitative summary of disease intensity over time, for comparison across in different week intervals. The AUDPC value was calculated by using the formula given in equation 2 by using the values of PDI in different week intervals. The AUDPC value is 2652.195. Similar study was done in onion and garlic by Abkhoo [14] and Shabnam and Suman [15] respectively. Nagesha and Nargund [13] also reported lower values of AUDPC in slow rust. In this study we made an effort to know the effect of weather condition in onion to causing purple blotch disease by doring the artificial spraying of inoculum of pathogen in open field condition to induction of diseases in plants. Results revelas that the delay in the sowing (November–December) increases the severity of onion purple blotch, reduces seed test weight and yield of crop.

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