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Management of early blight of potato through fungicides under field and laboratory conditions

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Abstract
Early blight caused by *Alternaria solani* (Ellis and Martin) Jones and Grout is one of the most serious diseases of potato throughout the world including India. Field experiments were conducted during *Rabi* season, 2016-17 and 2017-18 at Benuria, Bolpur under Red and Lateritic Agro-climatic zone of West Bengal for developing suitable management practice against early blight of potato (cv. Kufri Jyoti). Necessary laboratory studies were done in the Department of Plant Protection, Palli-Siksha Bhavana, Visva-Bharati. Symptoms of the disease appeared mainly on leaves, petioles and stems, and in severe condition on tubers. Initially the symptoms observed on lower most leaves as small, oval to irregular, dark brown to blackish spots that become enlarged with concentric rings. On microscopy, brownish muriform (9-11 transverse septa and 2-3 longitudinal septa) conidia in chains having almost hyaline beak were recorded that measured about 89.51 - 254.23 µm × 12.04 - 28.64 µm with an average size of 139.71 µm × 18.04 µm. All the fungicides tested (250, 500 and 1000 ppm concentrations) were effective against the pathogen when evaluated through poison food techniques in laboratory. Effectiveness of the fungicides was increased with the increase doses of fungicides. Indofil M-45 (96.30%) revealed superior where maximum inhibition of mean radial growth of the fungus was observed followed by Indofil Z-78 (94.44%), Merger (86.67%), Ishaan (84.44%), Foliogold (77.41%) and Infield Ayur (75.55%). Ethaboxam recorded ineffective and showed minimum (32.96%) inhibition. Fungicides used in field experiment were more or less efficacious. The highest disease reduction was recorded in Indofil M-45 (43.76%) followed by Indofil Z-78 (40.59%) and Merger (36.65%) over control whereas lowest of that was recorded in Ethaboxam (22.29%) and Infield Ayur (26.99%).

Keywords: *Alternaria solani*, early blight, potato, fungicides, management

1. Introduction
Potato (*Solanum tuberosum* L., Solanaceae) is the world's fourth most important food crop in terms of human consumption after rice, wheat and maize (Shekhawat, 2001; Bowen, 2003) [17, 1]. Potato is grown in almost all the states and union territories of India. The *Rabi* potato area is 614.78 thousand hectors in Uttar Pradesh followed by 405.00 thousand hectors in West Bengal and 321.25 thousand hectors in Bihar. Uttar Pradesh is the leading potato growing state in the country with a production of 15561.85 thousand MT followed by West Bengal (11052.60 thousand MT) and Bihar (6377.71 thousand MT) (Horticulture Statistics Division, 2018) [7].

The crop is suffering from numbers of diseases caused by fungi, bacteria, virus, viroid and phytoplasma (De, 2004) [4]. Early blight of potato caused by the fungus *Alternaria solani* is one of the most important foliar disease of potato worldwide and causing huge damage to the crop (Christ, 1990; Shtienberg *et al.*, 1990; Vander-Walls *et al.*, 2001) [3, 18, 19] and recognized as a threat since its first occurrence in Wisconsin in 1892 (Jones, 1893 and Rands, 1917) [9, 16]. In India, *A. solani* on leaves of potato was first reported from Farukhabad (Uttar Pradesh) by Butler in 1903 (Butler and Bisby, 1931) [2]. The disease primarily appears on leaves, stems, petioles, twigs and tubers as small, dark, dry, papery flecks, which grow to become brown-black, circular-to-oval areas with concentric rings on older leaves that favoured by warm temperatures and high humidity resulting in Defoliation, drying-off of twigs and thus causing loss from 50 to 86 per cent in tuber yield (Mathur and Shekhawat, 1986) [10]. At present no true resistant sources of the potato is available in the country, so chemical management is an indispensable component of Integrated Disease Management (IDM) programme. New fungicides are introducing in the country every year for controlling fungal diseases of crops. Therefore, the present study was undertaken to find out suitable management option especially chemical to combat the disease in both field and laboratory conditions.

2. Materials and Methods
2.1 The experimental site
The field experiment was conducted at the Farmers' field situated at Benuria, Birbhum near PSB Agricultural Farm, Visva-Bharati, Sriniketan, West Bengal. The experimental site is situated at an average altitude of 58.9 meter above MSL and 23º39′N latitude and 87º 42′E longitudes under Red and Lateritic Agro-climatic zones of West Bengal. The long-term average maximum temperature varied from 25°C to 30°C and long-term average minimum temperature varied from 10°C to 15°C in this region. The rainfall during the winter season was very low and sporadic, and daily sunshine hours were about 8 hours. The soil of the experimental site was slightly acidic (pH 5.5-6.0), sandy loam in texture having medium fertility status with good drainage facility.

2.2 Laboratory study

The laboratory experiment was conducted at Department of Plant Protection, Palli-Siksha Bhavana (Institute of Agriculture), Visva-Bharati, Sriniketan, West Bengal. The inhibitory effect of different concentrations (250, 500 and 1000 ppm) of seven fungicides (Table 1) was evaluated through poison food technique measuring per cent inhibition of mycelial growth of *A. solani* in *in-vitro* condition. Each treatment was replicated thrice following CRD. Potato dextrose agar was used as growth medium for the study. Each dose of fungicide was mixed thoroughly with the medium, poured into petridishes and allowed for solidification. The actively growing periphery of the nine-day old culture of *A. solani* was carefully removed using cork borer and transferred aseptically to the centre of each petridish containing the poisoned solid medium replacing the similar portion. Suitable control was maintained by growing the cultures on PDA without adding any fungicides. The plates were incubated at 27±1°C for nine days and the colony diameter (mm) was recorded at regular interval. When in control plate attained full of fungal growth, per cent inhibition of mycelial growth was calculated using the following formula (Vincent, 1947) [20]:

$$I = \frac{C - T}{C} \times 100$$

Where, I = per cent inhibition; C = growth in control; T = growth in treatment.

Characterization of the pathogen was made through colony morphology, vegetative and asexual reproductive structures produced by the fungus. Some small bits of infected leaf tissue were taken and placed in sterile petridish containing water, the pathogen grows beyond the area of the leaf tissue and produce good mycelial growth in water leading to formation of conidia. Microscopic observation is possible by placing such Petridish under microscope (Mondal *et al.*, 2015) [13].

2.3 Management of the disease in field condition

The field experiments on potato (cv. Kufri Jyoti) were set up during *Rabi* season, 2016-17 and 2017-18 following RBD with three replications and eight treatments including untreated control (Table 1). Seed tubers were treated with Indofil M–45 (Mancozeb 75% WP) @ 2g/kg before planting. General agronomic practices were followed to raise the crop. Spray solutions were prepared by dissolving specific amount of the chemicals in definite quantity of plain water. Spray was initiated just after detection of the disease symptoms in the experimental field, and repeated twice at an interval of 15 days.

Table 1: Fungicides used for the field and laboratory experiment against *A. solani*

Sl. No.	Fungicides	Active ingredient and formulation
1.	Indofil M-45 (Indofil Industries Ltd.)	Mancozeb 75% WP
2.	Foliogold (Syngenta India Ltd.)	Chlorothalonil 33% + Metalaxyl 3.3% SC
3.	Ethaboxam (Sumitomo Chemical India Ltd.)	Ethaboxam 40% SC
4.	Ishaan (Rallis Tata Enterprise)	Chlorothalonil 75% WP
5.	Indofil Z-78 (Indofil Industries Ltd.)	Zineb 75% WP
6.	Merger (Indofil Industries Ltd.)	Tricyclazole 18% + Mencozeb 62%
7.	Infield Ayur (Infield Organics Ltd.)	Eugenol 00.10% + Potassium salt of fatty acids 02.00% + Sodium salts 97.90% W/W

The incidence of early blight was recorded every seven days after each spraying starting from the first appearance of the disease. Twenty-five plants in each replicated plot were selected randomly and tagged. The whole plant was observed for scoring of disease severity. A descriptive disease rating scale (Horsefall and Barret, 1945) [6] was used for the assessment of the disease intensity (Table 2). Per cent disease index (PDI) was calculated by the following formula (McKinney and Davis, 1925) [11]:

$$PDI = \frac{\sum \text{Numerical ratings}}{\text{Total numbers of leaves observed} \times \text{maximum disease rating}} \times 100$$

The two years pooled data were subjected to statistical analysis after necessary transformation as per Panse and Sukhatme (1985) [15].

Table 2: Rating scale for assessment of early blight of potato

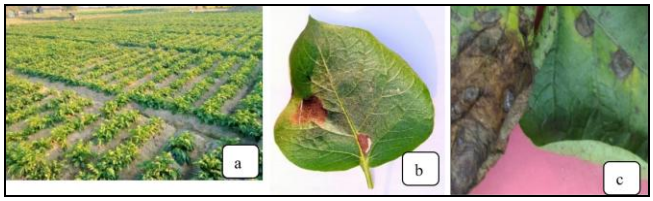
Category	Grade/ Numerical Rating	Description of the symptoms
I	0	Leaves free from infection.
II	1	Small irregular spots covering 1-10% leaf area.
III	2	Small irregular brown spots with concentric rings covering 11-25% leaf area.
IV	3	Lesions enlarged to form irregular brown with concentric rings covering 26-50% leaf area.
V	4	Lesions coalease to form irregular and appears as a typical blight symptom covering 51-75% leaf area.
VI	5	Lesions coalease to form irregular and appears as a typical blight symptom covering >76% leaf area.

3. Results and Discussion

3.1 Symptomatology

Symptoms appeared mainly on leaves, petioles and stems, and in severe condition on tubers. Initial symptoms were observed on lower most leaves i.e. older leaves, which consist of small, oval to irregular, dark brown to black, single to numerous necrotic spots. These spots become enlarged to form concentric rings (target-board like appearance). In most of the cases, a narrow, yellow halo was recorded surrounding the enlarged spot (Plate 1b and 1c). severely infected leaves dried out in and become papery (Plate 1c). Lesions in tubers were slightly sunken, dark brown to blackish and irregular than adjacent healthy skin. In some cases, a well-defined slightly raised margin between healthy and diseased tissue was observed. A dry rotting type symptom were recorded internally just beneath

the skin. Infected plants produced smaller tubers. The symptoms were more or less similar with the symptoms recorded by Middya (2017) [12], who stated that enlarged lesions that often delimited from healthy tissue by a narrow chlorotic halo due to toxins produced by the pathogen, which move in advance into uninfected epidermal cells.

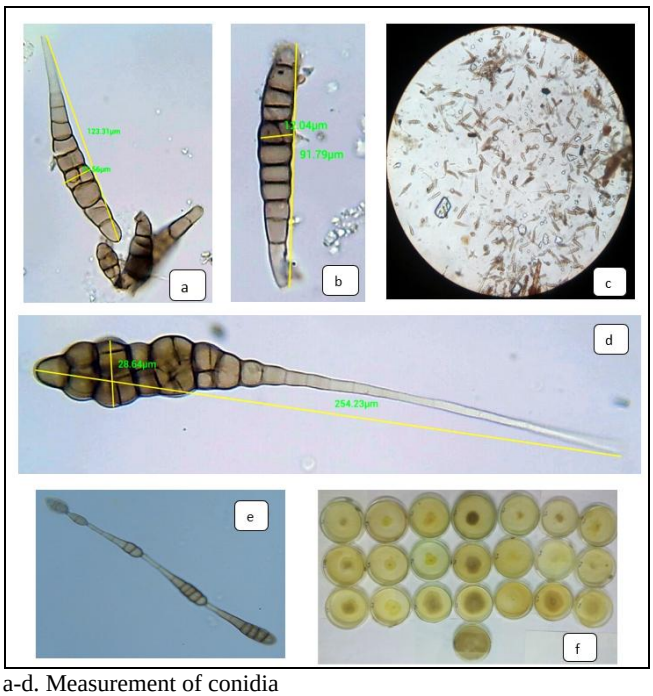


Experimental field
Appearance of typical concentric ring on leaf
Infected leaf showing chlorotic halo and drying out of leaf in severe infection

Plate 1: Symptoms of early blight of potato

3.2 Characterization of the pathogen

Colonies on PDA medium were initially whitish and hairy that became grey brown to black with velvety and cottony appearance. The pathogen produced septate and whitish mycelial growth on artificial medium (PDA medium) that turned into grey and black in time. Conidiophores were dark-brown, straight to slightly bent, septate and mostly solitary. In aqueous medium, the pathogen produced brownish coloured conidia in chains on conidiophores were muriform (9-11 transverse septa and 2-3 longitudinal septa) having beak (almost colourless), and size varied from 89.51 – 254.23µm × 12.04 – 28.64µm with an average size of 139.71µm × 18.04µm (Plate 2).



a-d. Measurement of conidia
e. Chain of conidia
f. Bio-assay of fungicides in in-vitro condition

Plate 2: Characteristics of the pathogen causing early blight

On the basis of morphological characters and comparison with the authentic description, the fungus was identified as *Alternaria solani* causing early blight of potato. The morphological descriptions of the pathogen almost corroborated with descriptions giving by Neergard (1945) [14]. According to Ellis (1971) [5], the solitary and beaked conidia have 9 to 11 transverse septa and a few or no longitudinal or oblique septa.

3.3 Laboratory screening of fungicides through poison food technique

The fungicides (Table 1) at different concentrations (250, 500 and 1000 ppm) were evaluated against *Alternaria solani* causing early blight of potato in laboratory by adopting poison food technique. The radial growth of the fungal mycelium was recorded at 7 days after inoculation (Table 3). Superiority of all the fungicides was recorded than the untreated control. Inhibition of mycelial growth recorded more with the increase of concentration of fungicides (Plate 2f). In all cases 1000ppm concentration gave maximum inhibition (Table 3). Maximum inhibition of radial growth (mean) was observed in Indofil M-45 (96.30%) followed by Indofil Z-78 (94.44%). These two fungicides can be categorized in Group-I, while Merger and Ishaan can be categorized in Group II. Inhibition of mean radial growth for Merger and Ishaan was 86.67% and 84.44%, respectively. Foliogold (77.41%) was also showed effective than Infield Ayur (75.55%). Ethaboxam was recorded ineffective against the pathogen causing early blight disease where lowest inhibition was observed (32.96%) (Table 3).

Table 3: *In-vitro* evaluation of fungicides against *A. solani*

Fungicides*	Per cent inhibition of mycelial growth			
	250 ppm	500 ppm	1000 ppm	Mean
T_1: Indofil M-45	88.89	100.00	100.00	96.30
T_2: Ishaan	73.33	84.44	95.56	84.44
T3: Foliogold	70.00	75.56	86.67	77.41
T_4: Indofil Z-78	83.33	100.00	100.00	94.44
T_5: Merger	81.11	85.56	93.33	86.67
T_6: Infield Ayur	64.44	72.22	90.00	75.55
T_7: Ethaboxam	30.00	32.22	36.67	32.96
SEm (±)	4.95	3.97	4.69	-
C.D. (p= 0.05)	14.60	11.72	13.82	-

*T_1= Indofil M-45 (Mancozeb 75% WP), T_2= Ishaan (Chlorothalonil 75% WP), T_3= FolioGold (Chlorothalonil 33% + Metalaxyl 3.3% SC), T_4= Indofil Z-78 (Zineb 75% WP), T_5= Merger (Tricyclazole 18% + Mencozeb 62%), T_6= Infield Ayur (Eugenol 00.10% + Potassium salt of fatty acids 02.00% + Sodium salts 97.90% W/W), T_7= Ethaboxam (Ethaboxam 40% SC), Radial growth of mycelium in control plates = 90mm

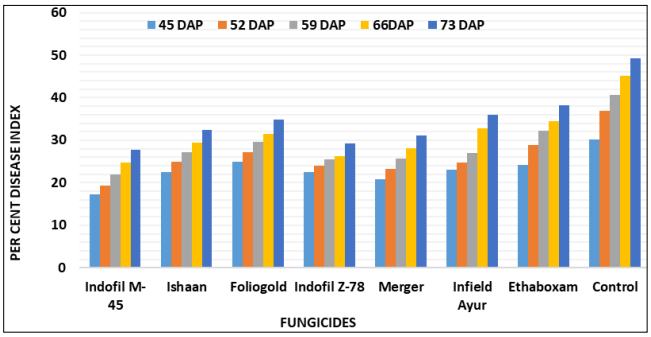


Fig 1: Efficacy of different fungicides against early blight of potato under field condition

3.4 Evaluation of fungicides in field condition

Experiment on management of early blight disease was carried out with different fungicides used (Table 1) in field condition (Plate 1a). The detailed procedure of experiments

has been mentioned under materials and methods. It is evident from the result presented in Table 4 that the treatment Mancozeb @ 2.5 g/l (T_1) exhibited best result in terms of disease intensity (27.69%) at 73 DAP as compared to others. No statistical difference was observed among the treatments, T_1 (Mancozeb), T_4 (Zineb), T_5 (Tricyclazole + Mancozeb) and T_2 (Chlorothalonil), where the disease intensity was recorded as 27.69%, 32.51%, 29.25% and 31.19%, respectively. In control (T_8), the disease intensity was 49.24%. No statistical differences were also observed among T_3 (Metalaxyl-M + Chlorothalonil) (34.77%), T_6 (Infield Ayur) (35.95%) and T_7 (Ethaboxam) (38.26%). Per cent reduction of disease over control was highest in T_1 (43.76) followed by T_4 (40.59) and T_5 (36.65). Not only that, the total tuber yield/plot was also highest in T_1 (24.68 t/ ha) followed by T_4 (23.34 t/ha), T_5 (22.72 t/ha) and T_2 (22.25 t/ha). No statistical difference was recorded between T_1 and T_4 in respect to yield/ha (Table 4). According to Islam *et al.* (2018) [8] Ethaboxam is highly effective against late blight of potato, while no satisfactory results of Ethaboxam on early blight were observed in this study.

4. Conclusion

Potato is one of the most important vegetable crops in India. Early blight disease of potato was prevalent in all potato growing areas in West Bengal. The study revealed that the disease can easily be identified by its characteristic's symptoms, and observing the asexual reproductive structures of the causal pathogen in aqueous medium that will help the farmers for selecting effective fungicide against the disease easily. Indofil M-45 exhibited most effective followed by Indofil Z-78 not only in reducing disease intensity but also increasing yield in the present study both in field and laboratory conditions. These fungicides are cost effective too. The results of the study can be recommended to the farmers for managing their potato fields against the disease by incorporating it into Integrated Disease Management (IDM) programme.

5. Conflict of interest

The authors declare that they have no conflict of interest.

6. Acknowledgement

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