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Tusi Chakma

Department of Plant Pathology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

PH Sobita Devi

Department of Plant Pathology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

Bireswar Sinha

Department of Plant Pathology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

Kota Chakrapani

Department of Plant Pathology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

N Gopi Mohan Singh

Department of Agricultural
Statistics, College of Agriculture,
Central Agricultural University,
Imphal, Manipur, India

TH Renuka Devi

Department of Genetics and
Plant Breeding, College of
Agriculture, Central Agricultural
University, Imphal, Manipur,
India

KH Ibohal Singh

Department of Entomology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

Corresponding Author:

Tusi Chakma

Department of Plant Pathology,
College of Agriculture, Central
Agricultural University, Imphal,
Manipur, India

In vitro studies on efficacy of plant extracts against *Alternaria solani* causing early blight of Potato

Tusi Chakma, PH Sobita Devi, Bireswar Sinha, Kota Chakrapani, N Gopi Mohan Singh, TH Renuka Devi and KH Ibohal Singh

Abstract

The efficacy of ten plant extracts and a check fungicide (Mancozeb) against *Alternaria solani* were evaluated at three different concentrations, *in vitro* by poisoned food technique to exploit their antifungal properties. Among the plant extract tested, the highest inhibition of 68.24%, 73.72% and 79.21% was shown by Garlic extract at 1%, 2% and 3% concentration followed by Adulsa, Turmeric, Ginger, Neem, Tulsi, Wild sedge, Marigold, Mentha, and Onion. Mancozeb at 0.1%, 0.2% and 0.3% concentrations showed 72.55%, 80.00% and 87.06% inhibition over control. Garlic (*Allium sativum* L.) was found most effective which exhibited maximum mycelial growth inhibition and minimum radial growth at higher concentration. Whereas, Onion extracts (*Allium cepa* L.) showed least mycelial growth inhibition (17.65%) and maximum radial growth (7.00 cm) at 20% concentration among the tested plant extracts.

Keywords: *Alternaria solani*, early blight, inhibition, plant extract, and potato

1. Introduction

Potato (*Solanum tuberosum* L.) is an important food crop of the world. Potato belongs to the family Solanaceae and also known as white or Irish potato. It is an important starchy food crop in sub-tropical and temperate regions. India ranks 4th in area under cultivation of potato and 3rd largest country in the world in production of potato and Uttar Pradesh is the largest potato producing state in the country and accounts for 31% of total production of potato. The top producers of potato in the world are India (28.01%), China (23.90%) and Russia (8.1%). Potato is grown almost in all the states of India. Major potato growing states are Uttar Pradesh, West Bengal, Bihar, Gujarat, Madhya Pradesh, Punjab, Assam, Chhattisgarh, Jharkhand and Haryana (Chakraborty *et al.*, 2022) [1]. Potato is a popular source of carbohydrate, and can be used both for table consumption as well as processed products. Raw potato contains 79% water, 17% carbohydrates (88% of which is starch), 2% protein and contains negligible fat. Potato plant is known to suffer from a number of fungal, bacterial and viral diseases and causes significant crop losses when favourable condition prevailed. Among the fungal diseases of potato, early blight caused by *Alternaria solani* is a major foliar disease and play a significant role in reducing the yield potentiality of the crop. However, primary damage by early blight is mainly attributed to premature defoliation of the potato plants, resulting in reduction of tuber yield. Average annual yield loss of potato due to this disease is approximately 75% of the total production depending upon the nature of the disease, weather conditions and type of cultivars (Dey and Chakraborty, 2012) [2]. In recent years, increase in *Alternaria solani* disease on potato foliage has been reported in various potato growing areas (Vloutoglou and Kalogerakis, 2000) [3]. Fungicides cannot be considered as a long-term solution, due to concerns of expense, exposure risks and the hazards of its residues. *Alternaria solani* has low sensitive with fungicides because of its production of dark brown to black pigment called melanin which enhanced survival and competitive abilities of the pathogen under certain environmental conditions (Bell and Wheeler, 1986) [4]. Moreover, the development of resistance of pathogenic fungi towards synthetic pesticides is a great problem that can significantly affect the efficacy of chemical fungicides (Kirk *et al.*, 2005) [5]. Plant extracts for the control of plant pathogens has emerged as a potential control strategy in recent years. Natural plant products are important sources of new agrochemicals for the control of plant diseases (Kagale *et al.*, 2004) [6]. Hence, the present investigation was undertaken to study the *in vitro* efficacy of aqueous plant extracts against *Alternaria solani* causing early blight of Potato.

2. Materials and Methods

2.1 Collection of the diseased samples and isolation of the causal pathogen: Potato leaves showing early blight symptoms were collected from farmer's fields and infected leaf samples were brought to laboratory and cut into small pieces measuring about 2 mm. Surface of specimen was sterilized with 0.1% sodium hypochlorite solution for 1 minute and washed thrice with sterilized distilled water. Then one piece of specimen was transferred on Potato Dextrose Agar (PDA) medium in the center of Petri plate and incubated at 27 ± 1 °C in BOD incubator and was observed periodically for the fungal growth. Pure culture of the *Alternaria solani* was obtained by hyphal tip culture technique and stored at 5 °C in refrigerator and periodically sub cultured to fresh medium throughout the experimental period.

2.2 In vitro efficacy of plant extracts against growth of *Alternaria solani*: Ten locally available plant extracts viz., Onion (*Allium cepa* L.), Ginger (*Zingiber officinales* L.), Mentha (*Mentha arvensis* L.), Wild sedge (*Lantana camara* L.), Turmeric (*Curcuma longa* L.), Marigold (*Tagetes erecta* L.), Garlic (*Allium sativum* L.), Neem (*Azadirachta indica* L.), Adulsa (*Justicia adhatoda* L.), Tulsi (*Ocimum tenuiflorum* L.) and a check fungicide (Mancozeb) was studied for their inhibitory effect against *Alternaria solani*. Fresh healthy plant parts were collected and washed with running tap water, followed by sterile water. These samples were air dried over a blotting paper. The air dried plant parts were crushed in mortar and pestle by adding sterile water at a ratio 1:1(w/v). The homogenate were filtered out through double layer of muslin cloth and filtrates were centrifuged at 1500 rpm for 15 minutes and the supernatants were collected. The resultant extracts were considered as 100% concentration. Each plant extracts were evaluated at three concentrations using poison food technique (Nene and Thapliyal, 1993) [7]. PDA was used as nutrient medium and required quantity of each plant extract and check fungicide was added separately to get a required concentration and sterilized in autoclave at 121 °C and 15 lb/inch² for 20 minutes. Twenty ml poisoned medium was poured to each of the 90 mm petri plates and five mm actively growing hyphal tip from periphery 7 days old culture of *Alternaria solani* was cut and transferred aseptically to the centre of each petri plate containing the poisoned medium/non-poison solid medium. Petri plates with only PDA (without plant extract) were served as control. The petri plates were incubated at 27 ± 1 °C and the radial growth of the fungus on the poisoned medium was recorded at time of mycelium growth reached 90 mm in control. Each set of treatment replicated three times. The percent inhibition of radial growth was calculated using formula described by Vincent (1947) [8].

Percent Inhibition of Radial Growth = $(C-T)/C \times 100$

Where, C= Radial growth of the fungus in control

T= Radial growth of the fungus in treatment

3. Results and Discussion

The experimental data on the efficacy of ten plant extracts and

a check fungicide on growth of *Alternaria solani* was evaluated by poisoned food technique *in vitro* and presented in Table 1 and Plate 1. Data revealed that plant extracts at different concentrations significantly reduced the radial growth of *Alternaria solani* over control and it was noticed that with the increase in the concentration of the extract, there was a corresponding increase in the inhibition of the pathogen.

Among the plant extract tested, Garlic extract showed maximum inhibition of 68.24%, 73.72% and 79.21% at 1%, 2% and 3% concentration and it was found significantly superior to other extracts. This was followed by Adulsa at 10%, 15% and 20% concentrations showed 46.67%, 55.29% and 62.35% of inhibition over control, respectively. Turmeric at 10% could inhibit 42.74% followed by Neem (34.12%), Ginger (32.16%), Wild sedge (31.76%), Tulsi (23.53%), Mentha (12.55%) and least inhibition of 8.23% was noticed in Marigold and Onion at 10% concentration. The result further revealed that Turmeric, Neem, Ginger, Wild sedge Tulsi, Mentha, Marigold and Onion at 15% concentration showed 48.63%, 41.18%, 37.65%, 35.29%, 31.76%, 17.65%, 14.90% and 10.98% of inhibition over control. However, at 20% concentration, Turmeric could inhibit 57.25% followed by Ginger (49.80%), Neem (47.06%), Tulsi (43.53%), Wild sedge (39.21%), Marigold (26.27%), Mentha (25.49%) and Onion (17.65%), respectively. Mancozeb at 0.1%, 0.2% and 0.3% concentrations showed 72.55%, 80.00% and 87.06% inhibition over control. The present study revealed that out of ten plant extracts, Garlic (*Allium sativum*) was found most effective which exhibited maximum mycelial growth inhibition (79.21%) and minimum radial growth (1.77 cm) at higher concentration. Whereas, the least mycelial growth inhibition (17.65%) and maximum radial growth (7.00 cm) was shown by Onion extracts at 20% concentration.

The result were in accordance with the findings of Devi *et al.*, (2017) [9] they reported among six aqueous plant extracts, *Allium sativum* at 1% concentration showed 86.2% inhibition whereas, 94.4% inhibition at 2% concentration and 100% inhibition was achieved at 3% and 4% concentrations. Plants naturally produce antifungal aromatic secondary metabolites, such as quinones, saponins, flavones, coumarins, flavonols, phenols, and tannins (Cowan, 1999) [10]. In the present study, the selected botanical extracts produced promising inhibition of *Alternaria solani*. So the effectiveness of Garlic extract in inhibition of mycelial growth of the pathogen might be due to presence of antimicrobial properties like diallyl-disulphide and diallyl-tri-sulphide in *Allium sativum*. This statement is in agreement with the findings of Kumar *et al.* (2018) [11] who reported that Garlic+Neem+Datura combination and only garlic extract induced 100% growth reduction at 10% and 20% concentrations for the test pathogen followed by Datura leaf extract at 10% and 20% concentration reduced the growth of *Alternaria solani* to 65.17% and 76.43%, respectively. The results are also supported by the findings of Ranaware *et al.* (2010) [12] they noticed that among seven plant extract tested Garlic bulb extract inhibited maximum growth of *Alternaria carthami* (48.68%) followed by Datura.

Table 1: Effect of plant extracts and a check fungicide on growth of *Alternari solani* in vitro

Sl. No.	Treatments	Plant parts used	Concentration (%)	Inhibition over control (%)*	Growth (cm)*
1.	Onion	Bulb	10	8.23 (0.92)	7.80
			15	10.98 (1.04)	7.57
			20	17.65 (1.25)	7.00
2.	Ginger	Rhizome	10	32.16 (1.51)	5.77
			15	37.65 (1.58)	5.30
			20	49.80 (1.70)	4.27
3.	Menthe	Leaf	10	12.55 (1.10)	7.43
			15	17.65 (1.25)	7.00
			20	25.49 (1.41)	6.33
4.	Wild sedge	Leaf	10	31.76 (1.50)	5.80
			15	35.29 (1.55)	5.50
			20	39.21 (1.59)	5.17
5.	Turmeric	Rhizome	10	42.74 (1.63)	4.87
			15	48.63 (1.69)	4.37
			20	57.25 (1.76)	3.63
6.	Marigold	Leaf	10	8.23 (0.92)	7.80
			15	14.90 (1.17)	7.23
			20	26.27 (1.42)	6.27
7.	Garlic	Clove	1	68.24 (1.83)	2.70
			3	73.72 (1.87)	2.23
			5	79.21 (1.90)	1.77
8.	Neem	Leaf	10	34.12 (1.53)	5.60
			15	41.18 (1.61)	5.00
			20	47.06 (1.67)	4.50
9.	Adulsa	leaf	10	46.67 (1.67)	4.53
			15	55.29 (1.74)	3.80
			20	62.35(1.79)	3.20
10.	Tulsi	Leaf	10	23.53 (1.37)	6.50
			15	31.76 (1.50)	5.80
			20	43.53 (1.64)	4.80
11.	Mancozeb	-	0.1	72.55 (1.86)	2.33
			0.2	80.00 (1.90)	1.70
			0.3	87.06 (1.94)	1.10
12.	Control	-	-	-	8.50
S.E(d)±			-	0.01	0.06
C.D. (5%)			-	0.02	0.12

* Mean of three replications, Values in parenthesis are log transformation

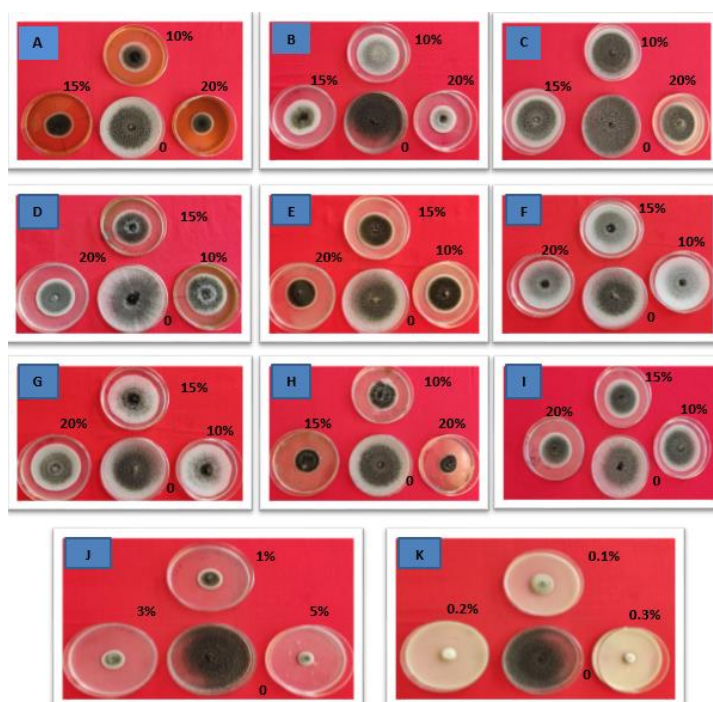


Plate 1: Effect of plant extracts on growth of *Alternari solani* in vitro at different concentration (A) Turmeric, (B) Mentha, (C) Marigold, (D) Wild sedge, (E) Ginger, (F) Onion, (G) Tulsi, (H) Adulsa, (I) Neem, (J) Garlic, (K) Mancozeb and (0) Control.

4. Conclusion

The results obtained from this study shows that among the plant extracts tested viz., Onion (*Allium cepa* L.), Ginger (*Zingiber officinales* L.), Mentha (*Mentha arvensis* L.), Wild sedge (*Lantana camara* L.), Turmeric (*Curcuma longa* L.), Marigold (*Tagetes erecta* L.), Garlic (*Allium sativum* L.), Neem (*Azadirachta indica* L.), Adulsa (*Justicia adhatoda* L.) and Tulsi (*Ocimum tenuiflorum* L.), Garlic (*Allium sativum*) showed the maximum mycelial growth inhibition of *Alternaria solani* under laboratory condition and so it can be used against early blight of potato. Therefore, use of plant extract could be suggested and recommended to be applied against *Alternaria solani* in field condition especially in order to reduce amount of chemicals and to reduce pollution hazards.

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