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Evaluation of efficacy of different botanicals and fungicides on *Alternaria solani*

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Abstract
Tomato is one of the most important crops for vegetable growers due to its increasing demand for consumption worldwide. To meet this requirement of demand and supply, its production needs to be enhanced. But early blight disease in tomato caused by *Alternaria solani* (Ellis and Martin) is a major concern, reducing yield to manifolds. In the current study, different plant extracts like Garlic (*Allium sativum*), Neem (*Azadirachta indica*), Datura (*Datura stramonium*), Tulsi (*Ocimum sanctum*), Ginger (*Zingiber officinale*) and Onion (*Allium cepa*) were tested in *in-vitro* condition, using food poison method and fungicides like Hexaconazole 5% SC at two different dose rates and Trifloxystrobin 25% + Tebuconazole 50% WG were tested in *in-vivo* condition, to understand its efficacy against *Alternaria solani* in Tomato. Among the tested botanicals, maximum percent inhibition of pathogen was observed in Datura (45.45%) followed by Ginger (38.12%) and Neem (33.80%). While amongst fungicide formulations, both the doses of hexaconazole 5% SC i.e. 640 and 800 ml ha^{-1} recorded better efficacy in controlling *A. solani* in tomato crop.

Keywords: Early blight, Alternaria solani, botanicals, fungicides, management, tomato

1. Introduction
Tomato (*Lycopersicon esculentum* Mill.) is one of the widely grown vegetables in the world and it is the second most important solanaceous vegetable crop. Tomato contains good source of antioxidants and Vitamin A & C which is necessary for metabolic activities for maintaining good human health. Worldwide it is cultivated in an area of 5.05 million ha with a production of 186.82 million tonnes and productivity of 36.97 tonnes ha^{-1} (Anonymous, 2020) [2]. In India, it is grown in a wide range of climatic condition across different states, accounting a total production of 21180.52 thousand tons (Anonymous, 2020) [2]. In Chhattisgarh, tomato is cultivated throughout the year with an acreage of 61.333 thousand ha, production and productivity of 1151.488 thousand tons and 18.77 tonnes ha^{-1} respectively (Anonymous, 2020-21) [3]. In Raipur district of Chhattisgarh, tomato is one of the most important crops for the vegetable growers. It is grown in an area of 4.130 thousand ha in Raipur district, with a production and productivity of 75.215 thousand tons and 18.21 tonnes ha^{-1} respectively. (Anonymous, 2020-21) [3].
During the entire growth period, tomato crop is exposed to several abiotic and biotic stresses. Amongst all, early blight of tomato caused by *Alternaria solani*, is most terrible disease, causing loss both at pre and post-harvest stages of the crop. This disease is identified by specific symptoms that initially occurs on older leaves of the plant and then spread to younger leaves. After sporulation, the initial symptoms emerge as large, dark brown to black spots that expand in size and stand out within concentric rings. This sporulation pattern results in the typical "target board" symptoms or bull eye-shaped spots. These concentric rings are very much visible when seen against the light. The suitable condition for sporulation of *A. solani* is 26 °C to 30 °C temperature, abundant moisture and humid environmental condition which is similar to the conditions prevailing in Raipur district of Chhattisgarh. Thus, the current study was initiated to understand the impact of different plant extracts and fungicides in controlling *Alternaria solani* in Tomato**.**

2. Materials and methods
2.1 Isolation and purification of *Alternaria solani*
The culture was purified by Single Spore Technique described by Kotasthane and Agrawal (2010). After the mycelial growth, in moist chamber petri plates, spores were studied with a stereoscopic microscope. The spores were then picked up, put to petri plates with simple water agar medium, and incubated at 25±2 °C for 24 hours.

These plates were examined for germinated spores under stereoscopic microscope, and then the spores were aseptically lifted by an inoculation needle and placed to PDA slants for further development.

2.2 Pathogenicity test of *Alternaria solani*

The pathogenicity test was conducted under open conditions in disposable glass using the attached pin prick method (Kiran *et al*., 2018 and Pathak *et al*., 2011) and in plastic petri plates using the detached pin prick method (Bandyopadhyay *et al*., 2016) to confirm the identification of early blight disease and its causal agent. Maintaining three replications for each isolates, tomato seeds were seeded in disposable glasses. For both attached and detached pin prick method, healthy plant leaves were selected and punctured with a thin needle to increase the infection. The PDA plates containing *A. solani* were mounted on pricked leaves after being cut with a 10 mm cork borer. To keep the culture moist, a wet cotton swab was placed on top of it. After symptom appearance on leaf, it was compared with the first symptoms.

2.3 *In-vitro* efficacy of botanicals on mycelial growth of *Alternaria solani*.

Tested the efficacy of plant extracts, against *Alternaria solani* using poisoned food technique under *in-vitro* conditions. Plant extracts of five botanicals *viz.* Garlic (*Allium sativum*), Neem (*Azadirachta indica*), Dhatura (*Datura stramonium*), Tulsi (*Ocimum sanctum*), Ginger (*Zingiber officinale*) and Onion (*Allium cepa*) were used for the test. For preparing the standard extract, 100 g of clean leaves or bulbs from each plant species were crushed using a mixture cum grinder and mixed in 100 ml of distilled water, which were then filtered through two layers of muslin cloth (Waghe *et al*., 2015) [14]. To test *Alternaria solani*, 10% of standard extract was taken along with PDA as basal culture medium in a conical flask and were mixed well. It was then autoclaved at 15 lbs psi pressure for 20 min. PDA (20 ml) was poured into 90 mm diameter petri plates and allowed to solidify. From a seven-day-old culture, a five-mm disc of *A. solani* was cut and positioned in the centre of petri dish. Three replications of each treatment were maintained and incubated at 25±2 ^{0}C.

2.4 *In-vivo* evaluation of fungicides against *Alternaria solani*

A field experiment was conducted in the Horticultural Instructional cum Research Farm, IGKV Raipur, to evaluate the efficacy of fungicides against *Alternaria solani*. Efficacy of fungicides were calculated using percent disease incidence, inhibition percentage.

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

Where,
I = Percent inhibition
C = Incidence in control
T = Incidence in treated plot (fungicides).

Treatment Details: Four treatments were considered *viz.*,
T1 = Hexaconazole 5% SC @ 640 ml ha^{-1}
T2 = Hexaconazole 5% SC @ 800 ml ha^{-1}
T3 = Trifloxystrobin 25% + Tebuconazole 50% WG @ 350 g ha^{-1}
T4 = Control

3. Results and Discussions

3.1 *In vitro* efficacy of plant extracts against *Alternaria solani*

An experiment was conducted to know the efficacy of different botanical extracts at 10% concentration and the results obtained have been summarized in Table 1. Out of the tested botanicals, Datura (*Datura stramonium*) was found to be highly effective in inhibiting the growth of *A. solani* with an inhibition of 45.45% which was followed by Ginger (*Zingiber officinale*), Neem (*Azadirachta indica*) and Garlic (*Allium sativum*) with an inhibition of 38.12%, 33.80% and 12.42% respectively. This might be due to the presence of higher percent of secondary metabolites in Datura.

The present findings are confirmed with the results of Ganie *et al*. (2013) [6], who reported superior control of *A. solani* by Datura (*Datura stramonium*). Similar other findings were reported by Nashwa and Abo-Elyousr (2012) [10], Waghe *et al*. (2015) [14], Raza *et al*. (2016) [11], Khalse *et al*. (2017) [8], Deshmukh *et al*. (2020) [5], Verma *et al*. (2020) [13].

Table 1: Efficacy of plant extracts against *Alternaria solani*

Botanicals	Mycelium diameter (mm)	Percent inhibition (%)
T1- Garlic	78.82	12.42
T2-Neem	59.58	33.80
T3-Datura	49.09	45.45
T4-Tulsi	84.71	5.88
T5-Ginger	55.69	38.12
T6-Onion	88.49	1.67
T7-Control	89.09	1.01
CV %	2.73	11.06
S.E(m) ±	1.14	1.26
CD at 1%	3.48	3.86

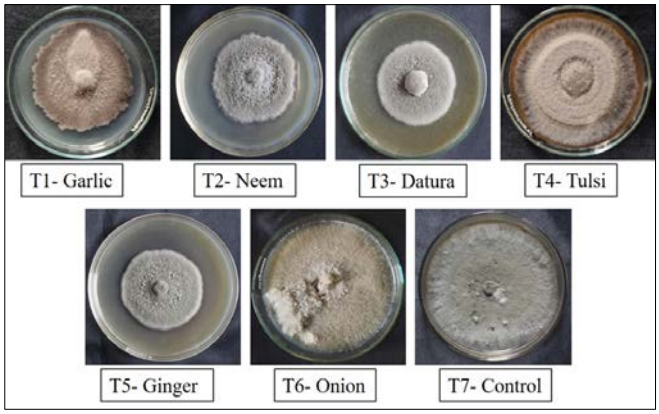

Fig 1: Effect of Botanicals on growth and colony characters of *A. solani*

3.2 *In-vivo* efficacy of fungicides against *Alternaria solani*

Efficacy of different fungicides was studied against early blight disease incidence on Pusa Ruby variety of tomato and results were illustrated in Table 2. The data on PDI of early blight was recorded after 65 and 95 days after transplanting (DAT) in each plot. Efficacy of fungicides was evaluated by using PDI %. The PDI showed the inhibition capacity of fungicides to control diseases in each treatment. It was observed that T1 (Hexaconazole 5% SC @ 640 ml ha^{-1}) recorded maximum efficacy (74.02%) followed by T2 (Hexaconazole 5% SC @ 800 ml ha^{-1}) with an efficacy of 73.81%, and they were statistically *at par* with each other. This might be due interference of Hexaconazole in the process of building fungal cell wall and finally inhibiting

further growth of the fungus. Yadav *et al.*, 2018 [16] also recorded similar results of Hexaconazole in *A. solani*.

Also, the considered fungicides like Hexaconazole 5% SC (Contaf Plus) and Trifloxystrobin 25% + Tebuconazole 50% WG (Nativo) did not record any visual phytotoxicity symptoms on tested tomato plants and hence can be considered safe on Tomato. Similar findings were also reported by Amin *et al.*, 2018 in Cumin crop and Varalakshmi *et al.*, 2020 [15] in grapes.

Table 2: Efficacy of fungicides against *Alternaria solani*

Treatment	Dose	PDI %	Inhibition (%)
T1- Hexaconazole 5% SC	640 ml ha^{-1}	21.00 (26.54)*	74.02 (60.21)
T2- Hexaconazole 5% SC	800 ml ha^{-1}	21.67 (27.08)	73.81 (59.86)
T3- Trifloxystrobin 25% + Tebuconazole 50% WG	350 g ha^{-1}	25.00 (28.44)	67.99 (57.31)
T4- Control		83.33 (66.61)	0.00
CV %		31.44	22.03
S.E(m) ±		6.85	6.86
CD at 5%		24.18	24.21

*Figures in parenthesis are angular transformed value

Fig 2: Efficacy of fungicides against *A. solani*

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