The Pharma Innovation Journal 2023; 12(2): 3794-3796

www.ThePharmaJournal.com

The Pharma Innovation

**ISSN (E): 2277-7695**
**ISSN (P): 2349-8242**
**NAAS Rating: 5.23**
TPI 2023; 12(2): 3794-3796

www.thepharmajournal.com
Received: 21-11-2022
Accepted: 24-12-2022

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# Efficacy of different fungicides, plant extracts and bio-agents against the *Alternaria solani* under *in vitro* conditions

**Bimla, RK Bagri and Lalita lakhran**

**Abstract**
S-22 tomato (*Solanum lycopersicum*) is a cultivated variety of tomato belonging to family Solanaceae. The crop S-22 tomato is infected by many diseases including fungi, bacteria and viruses. Among the fungus diseases, Early blight of tomato caused by *Alternaria solani* during all stages of plant growth. In the experiment of *in vitro* efficacy of bio-control agents against *Alternaria solani* all treatments showed significant effect to inhibit growth of the pathogen. Maximum mycelial growth inhibition was observed with *Allium sativum* clove extracts followed by *Azadirachta indica, Gingiber officinali, Ocimum sanctum, Calotropis gigantia, Datura stramonium* and *Aloe barbadensis* leaf extracts. Percent mycelia growth inhibition of disease organism was ranged from 35.84 to 83.15 percent.

**Keywords:** Tomato, *Alternaria solani*, *in-vitro* and plant extract

## Introduction
Tomato (*Lycopersicon esculantum* Mill.) is the second most important vegetable crop after potato, which belongs to the family *solanaceae*. Tomato is a native to Peruvian and Mexican region. It is a well-known fact that tomato is a main fruit consumed as a vegetable globally which provides important minerals, vitamins, fibres and antioxidants. Tomato is grown for its edible fruits, which can be consumed either fresh or in the form of various processed products. Tomato has high medicinal value, its pulp and juice is digestible promoter of gastric secretion and blood purifier. China is the leading country in production of tomato (31%), followed by India and the United States with the second and third highest producer in the world. The consumption of tomato stands second after potato being rich in vitamins (K, C and A), minerals (Fe, Ca and P), amino acids, sugars, dietary fibres and antioxidant and contains 95.3% of water [Gomes *et al*, 2010, Awan *et al*., 2019,] [5, 3]. In India tomato production is 21055.85 million tonnes and area 865.29 Million ha (Anonymous 2021,) [1]. In Rajasthan, It occupied an area of 20.50 Million ha with an annual production of 232.86Mt (Anonymous 2021) [1]. Tomato plants are suffered with large number of biotic stresses including insect pests and diseases from the time of emergence to harvest. It suffers with various diseases incited by fungi, bacteria, viruses, nematodes etc. in several countries (Mark and Brooke 2006) [7]. It is highly sensitive to abiotic stresses especially extreme temperature, salinity, drought, excessive moisture and environmental pollution and biotic stresses. More than 200 diseases have been reported to infect tomato in the world (Atherton and Rudich, 1986) [2]. Among the fungal diseases, early blight also known as target spot disease incited by *Alternaria solani*. It is very difficult to manage Alternaria blight because the pathogen has wide host range, extreme variability in pathogenic isolates and prolonged active phase of the disease cycle. The causal organism is air borne and soil inhabiting and is responsible for early blight, collar rot and fruit rot of tomato (Datar and Mayee, 1981) [4]. In India it causes about 72% of total production loss every year and about 1.36% of yield loss every year (Gomes *et al*., 2010) [5].
This disease is very difficult to control (Pasche *et al.*, 2005) [8]. Failure to control this disease can cause reduction in yield (Malik *et al*., 2014) [10]. There is need to evaluate the efficacy of new effective fungicides and bioagents which will be helpful in increasing the quality and quantity of tomato production (Sahu *et al*., 2013) [13]. Including these techniques for the management of early blight in tomato also the use of various botanicals have been recommended by various researchers. These include onions, ginger, garlic and neem plants for the extraction of various types of botanicals (Swami *et al*., 2013) [14]. The use of neem leaves have been very prominent among all the botanicals. Various fungicides *viz*., Chlorthalonil,

Mancozeb, Hexaconazole and Azoxystrobin when used at different concentrations *viz*., 1000, 500, 200, 100, 50 ppm under field conditions proved to be very effective against this pathogen and helped to reduce its severity in various crops (Prasad and Naik, 2003) [12].

## Materials and Methods

### Efficacy of plant extracts against *Alternaria solani in vitro* conditions

The activity of plant extracts were evaluated using poisoned food technique (*in vitro*) at different concentrations *viz*., 500, 1000 and 1500 ppm. Tested efficacy of plant extracts against *Alternaria solani* using poisoned food technique under *in vitro* conditions (Nene and Thapliyal, 1993) [11]. Fresh healthy plant parts of 100 g (leaves/fruit/bulb) were collected from field, then they were washed with tap water, air dried and crushed in 100 ml of sterile water. Potato dextrose agar medium was used as nutrient medium and required quantity of each plant extract was added separately to get a required concentration of the plant extract. The plant extract were thoroughly mixed with PDA medium and sterilized at 121 °C for 20 minutes. Twenty milliliter of poisoned medium was poured to each of the 90 mm Petri dish and allowed for solidification. Simultaneously without plant extract PDA was poured in Petri dishes as control. Actively growing periphery of the 7- day- old culture of *A. solani* was carefully cut using a cork borer and transferred aseptically to the centre of each petri dish containing the poisoned/non-poison solid medium. The plates were incubated at 25±2 °C. Each treatment was replicated three times.

## Result and Discussion

### Efficacy of plant extracts against *Alternaria solani* by poisoned food technique

The aqueous extracts of seven plant species (leaf/clove/ rhizomes) were tested at three concentrations (each @ 500, 1000 and 1500 ppm) to know their bio efficacy against *Alternaria solani* using poisoned food technique under *in vitro* condition on Potato Dextrose Agar medium.

Efficacy of plant extracts against pathogen showed that all the tested plant extracts numerically inhibited mycelial growth of *Alternaria solani*, contrast to untreated control. The percent mycelial growth inhibition of *Alternaria solani* was increased with increased in concentration from 500 to 1500 ppm of plant extracts.

At 500 ppm concentration, mycelial growth inhibition of pathogen ranged from 28.92 to 78.62 percent. However maximum mycelial growth inhibition was recorded with *Allium sativum* cloves extract (78.62%) which was found significantly superior over rest of tested plant extracts. This was followed by *Azadirachta indica* leaf extracts with 76.62 percent mycelial growth inhibition and also found significantly second highest mycelial growth inhibition after *Allium sativum* clove extract. Whereas *Gingiber officinali*, *Ocimum sanctum*, *Calotropis gigantia* and *Datura stramonium* leaf extracts recorded third, fourth, fifth and sixth maximum mycelial growth inhibition with 70.66, 35.98, 32.81 and 30.69 percent respectively. Lowest mycelial growth inhibition of 28.92 percent was observed with *Aloe barbadensis* leaf extracts.

Efficacy of seven plant extracts against *Alternaria solani* at 1000 ppm concentration showed that increased concentration of plant extracts increased the mycelial growth inhibition. All the plant extracts showed mycelial growth inhibition in the range of 35.66 to 84.14 percent inhibition. Highest mycelial growth inhibition was observed in case of *Allium sativum* clove extract with 84.14 percent inhibition which was found statistically significant than the other plant extracts. However, *Azadirachta indica* and *Gingiber officinali* extract recorded 80.09 and 75.45 percent mycelial growth inhibition which found second and third after *Allium sativum* clove extract for inhibition. Whereas *Ocimum sanctum*, *Calotropis gigantia* and *Datura stramonium* leaf extracst recorded 48.04, 44.69 and 39.92 percent mycelial growth inhibition respectively in desending order. Plant extracts, *Aloe barbadensis* leaf extract showed least mycelial growth inhibition (35.66%). At 1000 ppm concentration, all the plant extracts tested effective and recorded higher mycelial growth inhibition as compared to 500 ppm concentration.

At 1500 ppm concentration, mycelial growth inhibition was observed higher than the 500 and 1000 ppm concentrations with the range of 42.95 to 86.69 percent inhibition. However, maximum mycelial growth inhibition was observed with *Allium sativum* clove extracts by 86.69 percent and found significantly superior to the other plant extracts followed by *Azadirachta indica*, *Gingiber officinali*, *Ocimum sanctum*, *Calotropis gigantia* and *Datura stramonium* leaf extracts inhibited mycelial growth in decreasing order with 81.96, 81.58, 52.55 51.05 and 45.66 percent. Least mycelial growth inhibition was recorded in *Aloe barbadensis* leaf extracts with 42.95 percent.

Based on all concentrations of plant extracts, maximum mycelial growth inhibition was observed with *Allium sativum* clove extracts followed by *Azadirachta indica*, *Gingiber officinali*, *Ocimum sanctum*, *Calotropis gigantia*, *Datura stramonium* and *Aloe barbadensis* leaf extracts. Data on mean percent mycelia growth inhibition of disease organism was ranged from 35.84 to 83.15 percent. Among the plant extracts, *Allium sativum* recorded highest mean mycelium growth inhibition of 83.15 percent which was found significantly superior over rest of the plant extracts followed by *Azadirachta indica* leaf extract with 79.56 percent, *Gingiber officinali* with 75.90 percent, *Ocimum sanctum* with 45.52 percent, *Calotropis gigantia* with 42.85 percent, *Datura stramonium* with 38.76 percent and *Aloe barbadensis* with 35.84 percent mean mycelial growth inhibition recorded in decreasing order. The overzealous and indiscriminate use of fungicides may cause environmental and toxicological problems. Recently, plant products are used as plant protectors because some plants have toxic compounds to kill pathogens. It is safely, eco-friendly and bio products. To avoid the use of synthetic fungicides, use of organic plant bio products, phenolic acid and flavonoids is gaining importance in plant disease control. Kumar *et al.* (2021) [9] in which they tested the efficacy of five locally available plant extracts for their antifungal activity against the early blight of potato incited by *Alternaria solani.* The extracts included *Datura stramonium*, *Allium sativum*, *Azadirachta indica*, *Eucalyptus globulus*, and *Lantana camara.* All extracts reduced mycelial growth and conidial germination of *A. solani. In vitro* studies showed that extracts obtained from *A. sativum* and *A. indica* have significant inhibition of mycelial growth of *A. solani* (88.80 and 86.62 percent) at 20 percent concentration. Higher concentrations of *A. sativum* extract caused a higher reduction of *A. solani* radial growth on potato dextrose agar medium. Extracts obtained from *A. sativum* and *A. indica* at 20 percent

concentration, were found most effective for inhibition of conidial germination of 85.50 and 80.04 percent respectively of *A. solani.*

**Table 1:** Efficacy of plant extracts against *Alternaria solani* by poisoned food technique

| S. No. | Plant extracts | Plant part used | Percent inhibition of mycelia growth at various concentration | | | Mean |
|---|---|---|---|---|---|---|
| | | | 500 ppm | 1000 ppm | 1500 ppm | |
| 1 | Aak | Leaves | 32.81 | 44.69 | 51.05 | 42.85 |
| | | | (34.95) | (41.95) | (45.60) | (40.89) |
| 2 | Tulsi | Leaves | 35.98 | 48.04 | 52.55 | 45.52 |
| | | | (36.86) | (43.88) | (46.46) | (42.43) |
| 3 | Ginger | Rhizomes | 70.66 | 75.45 | 81.58 | 75.90 |
| | | | (57.20) | (60.30) | (64.58) | (60.60) |
| 4 | Datura | Leaves | 30.69 | 39.92 | 45.66 | 38.76 |
| | | | (33.64) | (39.18) | (42.51) | (38.50) |
| 5 | Neem | Leaves | 76.62 | 80.09 | 81.96 | 79.56 |
| | | | (61.08) | (63.50) | (64.87) | (63.12) |
| 6 | Garlic | Cloves | 78.62 | 84.14 | 86.69 | 83.15 |
| | | | (62.46) | (66.53) | (68.60) | (65.76) |
| 7 | Aloevera | Leaves | 28.92 | 35.66 | 42.95 | 35.84 |
| | | | (32.53) | (36.67) | (40.95) | (36.78) |
| 8 | Control | | 0.00 | 0.00 | 0.00 | |
| | | | (0.00) | (0.00) | (0.00) | |
| | | | S.Em± | CD (P=0.05) | | |
| | | P | 0.19 | 0.52 | | |
| | | C | 0.11 | 0.32 | | |
| | | PxC | 0.32 | 0.90 | | |

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