*Plant Archives* Vol. 17 No. 1, 2017 pp. 111-114 *ISSN 0972-5210*

# *IN VITRO* EVALUATION OF FUNGICIDES AGAINST *CERATOCYSTIS FIMBRIATA* ELL. AND HALST. CAUSING WILT IN POMEGRANATE

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## Abstract

Pomegranate (*Punica granatum* L.) is an ancient fruit, belonging to the smallest botanical family punicaceae and pomegranate is a native of Iran. It is commercially an important fruit crop of both tropical and subtropical regions. In India, it is regarded as a "vital cash crop", grown in an area of 1, 16,000 ha with a production of 89,000 MT with an average productivity of 7.3 MT. Karnataka State has the distribution of cultivating pomegranate under tropical condition in an area of 12,042 ha with a production of 1, 29, 547 tonnes. Out of nine fungicides tested, carbendazim, hexaconazole, thiophanate methyl, propiconazole and tebuconazole showed 100 per cent inhibition at all concentrations (0.05%, 0.10% and 0.15%, respectively).

***Key words :*** Pomegranate, wilt, *Ceratocystis fimbriata*, plant and per cent.

## Introduction

Pomegranate (*Punica granatum* L.) is an ancient fruit, belonging to the smallest botanical family punicaceae and pomegranate is a native of Iran. It is commercially an important fruit crop of both tropical and subtropical regions. In India, it is regarded as a "vital cash crop", grown in an area of 1,16,000 ha with a production of 89,000 MT with an average productivity of 7.3 MT. Karnataka State has the distribution of cultivating pomegranate under tropical condition in an area of 12,042 ha with a production of 1,29,547 tonnes. Where this crop has spread across different districts *viz*., Bijapur, Bellary, Bagalkot, Koppal, Chitradurga, Belgaum, Davangere, Tumkur, Bangalore and Gulbarga. Pomegranate suffers from ten economically important diseases, among them bacterial blight or spot, fruit rot, anthracnose and wilt complex are severe and cause significant losses in recent years. Wilt caused by *Ceratocystis fimbriata* is the most severe disease in Karnataka, which causes yellowing, drooping and death of pomegranate plant leading to loss to the farmers. It is pertinent to generate information on the efficacy of available and new fungicides *in vitro* to manage wilt within the reasonable limit of fungicidal residues permit by the importing countries.

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## Materials and Methods

### Isolation of the pathogen

*Ceratocystis fimbriata*, associated with wilt was isolated from the infected stems and roots of pomegranate plant which were collected from Ganjalli field. The sliced pieces of collected stem portions with characteristic symptoms of vascular staining were surface sterilized with 1 per cent $NaHCO_3$ (sodium hypochlorite) for about 2 minutes and washed in alcohol (70%) and twice with sterile water to remove traces of $NaHCO_3$. Pathogen isolation was made using carrot bait technique (Moller and DeVay, 1968) in which, stems were placed in between the carrot disks and kept in a humid chamber and incubated at 25±2°C under 12 hour photoperiod (Moller and DeVay, 1968). After perithecium formation, a portion of the fungi was transferred to freshly prepared PDA and oat meal agar media to allow the full development of fungi. In order to confirm the identity of the fungus, the ascospores, aleroconidia, endoconidia and perithecia were observed under the high power (40x) microscope from Raichur isolates the pure culture. The identification of studies of pathogen has done as explained by Sharma *et al*. (2010).

### *In-vitro* evaluation of fungicides

*In vitro* evaluation of commonly available fungicide molecules was carried out in separate set of experiments with completely randomized design using poisoned food method against *Ceratocystis fimbriata.*

In poisoned food method, 20 ml of oat meal agar medium was initially mixed with fungicides was poured in to 90 mm diameter petri dishes and control treatment was maintained without addition of fungicides. After solidification, 5 mm discs of *C. fimbriata* was placed at the centre of the plate. Each set of experiment was replicated four times and plates were incubated at 26±2°C. When the growth of test fungus reached periphery of petriplate in the control treatment, observations were taken on colony diameter. Later, per cent inhibition of growth was calculated using the formula (Vincent, 1927).

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

Where,

I = Per cent inhibition.

C = Radial growth of fungus in control.

T = Radial growth of fungus in treatment.

## Results and Discussion

Nine fungicides were tested against *C. fimbriata* by poisoned food technique. Among them, carbendazim, hexaconazole, thiophanate methyl, propiconazole and tebuconazole showed 100 per cent inhibition at (0.05%, 0.10% and 0.15%) all the concentrations. However, inhibition of 96.3 per cent was in difenconazole and tricyclazole (91.03%). Least inhibition was found in azoxystrobin with 48.90 per cent, 65.70 per cent and 80.30 per cent at 0.05 per cent, 0.10 per cent and 0.15 per cent concentrations, respectively (table 1 and plate 1). *In vitro* evaluation of fungicides provides preliminary information regarding its efficacy against a pathogen with in a shortest period of time and therefore serve as guide for further field testing. In the present study, In the present investigation, fungicides *viz.,* carbendazim, hexaconazole, thiophanate methyl, propiconazole and tebuconazole showed cent per cent inhibition at all concentrations (0.05%, 0.10% and 0.15%) and tricyclazole showed 73.11 per cent inhibition at 0.05 per cent concentration, 100 per cent inhibition at 0.10 per cent and 0.15 per cent concentrations (fig. 1). The efficacy of the triazoles fungicides such as hexaconazole and tricyclazole may be attributed to their interference with the biosynthesis of fungal sterols and inhibition of ergosterol biosynthesis. In many fungi, ergosterol is essential to the structure of cell wall and its absence causes irreparable damage to

**Table 1 :** *In vitro* evaluation of different fungicides against *C. fimbriata*.

| S. no. | Fungicide | Per cent inhibition at different concentrations (%) | | | |
|---|---|---|---|---|---|
| | | 0.05 | 0.10 | 0.15 | Mean |
| 1. | Carbendazim | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) | 100.0(90.0)* |
| 2. | Triadimefon | 89.0(70.6) | 95.5(77.8) | 97.3(80.5) | 93.9(75.7) |
| 3. | Hexaconazole | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) |
| 4. | Thiophanate methyl | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) |
| 5. | Difenoconazole | 95.3(77.5) | 95.7(78.0) | 97.9(81.7) | 96.3(78.9) |
| 6. | Propiconazole | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) |
| 7. | Azoxystrobin | 48.9(44.4) | 65.7(54.2) | 80.3(63.7) | 65.0(53.7) |
| 8. | Tricyclazole | 73.11(58.8) | 100.0(90.0) | 100.0(90.0) | 91.03(72.57) |
| 9. | Tebuconazole | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) | 100.0(90.0) |
| | **Mean** | 80.63(63.9) | 85.69(67.7) | 87.55(69.3) | 84.62(66.9) |

| | S. Em. ± | CD at 1% |
|---|---|---|
| **Fungicides (F)** | 0.10 | 0.37 |
| **Concentration (C)** | 0.05 | 0.20 |
| **F × C** | 0.17 | 0.64 |

* Figures in parenthesis arc sine transformed value.

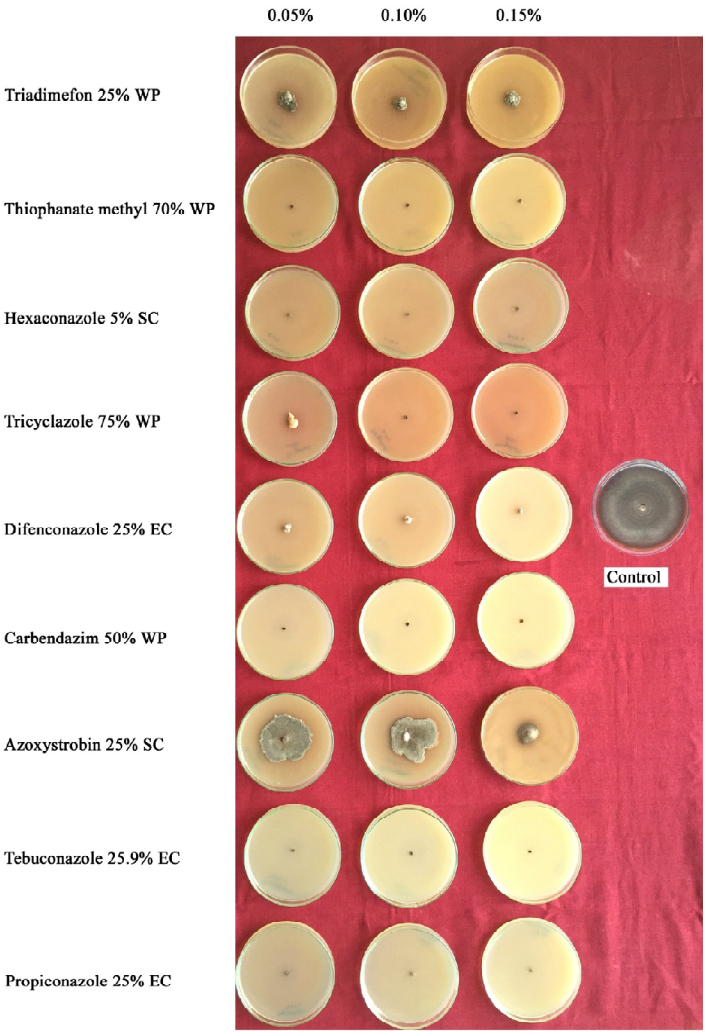


**Plate 1 :** *In vitro* evaluation of different fungicides against *Ceratocystis fimbriata*.

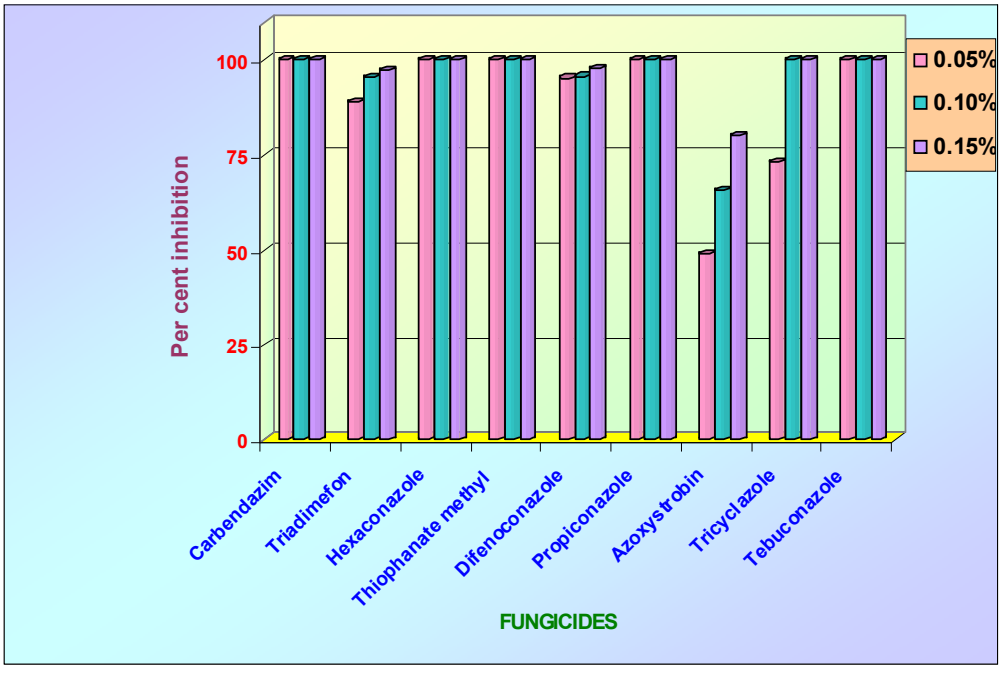


**Fig. 1 :** *In vitro* evaluation of different fungicides against *C. fimbriata*.

cell wall leading to death of fungal cell. A similar study was reported for the effectiveness of triazoles, which inhibit the biosynthesis pathway in fungi (Nene and Thapliyal, 1973). These findings are supported by earlier workers (Vijaya *et al*., 2006; Somasekhara, 2009; Khosla, 2013; Sonyal *et al*., 2016 and Chaudhari *et al*., 2016)

## References

Chaudhari, V. G., Priyanka Kshirsagar and A. M. Tirmali (2016). Studies on wilt complex disease of pomegranate (*Punica granatum* L.). *Advances in Life Sciences*, **5(3)** : 747-755.

Khosla, K. (2013). Evaluation of fungicides and plant extracts against *Ceratocystis fimbriata* causing wilt of pomegranate. *J. Mycol. Pl. Pathol*., **43(2)** : 193-197.

Moller, W. J. and J. E. Devay (1968). Carrot as a species selective isolation medium for *Ceratocystis fimbriata*. *Phytopathology*, **58** : 123.

Nene, Y. L. and P. N. Thapliyal (1973). *Fungicides in Plant Disease Control*. Oxford and IBH publishing house, New Delhi. pp. 163.

Sharma, K. K., J. Sharma and V. T. Jadhav (2010). Etiology of pomegranate wilt and its management. In: *Fruit, Vegetable, Cereal science and Biotechnology* 4 (special issue **2**), Global Science Books, 96-101.

Somasekhara, Y. M. (2009). *In vitro* and *in vivo* evaluation of fungicides for the management of pomegranate wilt pathogen (*Ceratocystis fimbriata*). Paper Presented in: 2nd *Inter. Symp. Pomegranate and minor including Mediterranean Fruits,* Univ. Agric. Sci., Dharwad, June 23-27, pp. 140-141.

Sonyal, S., Madhu, H. S. Mahesh, B. Palanna, S. H. Manjunath and A. Papachan (2016). Effect of fungicides on growth of *Ceratocystis fimbriata* Ell. and Halst. causing wilt in pomegranate. *Int. J. Pure App. Biosci*., **3(4)** : 28-32.

Vijaya, H. K., K. Srikant and R. H. Yashoda (2006). Chemical control of sett rot of sugar cane caused by *Ceratocystis paradoxa*. *Karnataka J. Agric. Sci*., **20(1)** : 62-64.

Vincent, J. M. (1927). Distortion of fungal hyphae in presence of certain inhibitors. *Nature*, **159** : 850.