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In vitro evaluation of combi-fungicides against *Ceratocystis fimbriata* causing wilt in pomegranate

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Abstract
Pomegranate (*Punica granatum* L.) is an old fruit crop, having a place with the littlest herbal family punicaceae and pomegranate is a local of Iran. It is financially a significant organic product harvest of both tropical and subtropical locales. In India, it is viewed as an "essential money crop", filled in a space of 1, 16,000 ha with a creation of 89,000 MT with a typical efficiency of 7.3 MT. Karnataka State has the dissemination of developing pomegranate under tropical conditions in a space of 12,042 ha with a creation of 1, 29, 547 tons. It is affected by several diseases of which wilt one of the most important diseases caused by *Ceratocystis fimbriata*. Seven combi-fungicide compounds were evaluated for effectiveness using the approach of poisoned food against *C. fimbriata* (Cf-26). Hexaconazole + zineb, carbendazim + mancozeb, trifloxystrobin + tebuconazole, and captan + hexaconazole were determined to be among the combi-fungicides that were most effective.

Keywords: Combi-fungicides**,** concentrations, *Ceratocystis fimbriata*, percent

Introduction
Pomegranate (*Punica granatum* L.) is a historic fruit that originates from Iran and is a member of the punicaceae, the smallest botanical family. It is a crucial fruit crop for both tropical and subtropical climates commercially. It is grown in 1,16,000 ha area in India and is said to as a "vital cash crop", since it produces 89,000 MT with an average yield of 7.3 MT. In the Karnataka area distribution of 12,042 ha and a yield of 1,29,547 tonnes of production (http://nhb.gov.in), the state of Karnataka is home to pomegranate cultivation. Bijapur, Bellary, Bagalkot, Koppal, Chitradurga, Belgaum, Davangere, Tumkur, Bangalore, and Gulbarga are among the areas where this crop has spread. Among the ten commercially significant diseases that affect pomegranates, bacterial blight or spot, fruit rot, anthracnose, and wilt complex are severe and have recently resulted in large losses. The most serious disease in Karnataka is wilt caused by *Ceratocystis fimbriata*, which kills pomegranate plants and causes yellowing, drooping, and death, costing farmers money. It is important to gather data on the effectiveness of novel and existing fungicides *in vitro* to control wilt within the acceptable range of fungicidal residues allowed by the importing nations.

Materials and Methods
Isolation of the pathogen: The wilt-related *Ceratocystis fimbriata* was found infected in the pomegranate plant stems and roots that were taken from the Ganjalli field and isolated the same. The sliced portions of the collected stem segments that displayed the vascular staining symptoms were surface sterilized with 1% sodium hypochlorite (NaHCO3) for about 2 minutes before being cleaned twice in sterile water and once in 70% alcohol to get rid of any remaining NaHCO3 traces. The carrot bait approach (Moller and DeVay,) [3] was used to isolate the pathogen. In this method, stems were inserted between carrot disks, stored in a humid chamber, and cultured at 25 °C with a 12-hour photoperiod. A part of the fungus was moved to freshly made PDA and oat meal agar media after perithecium formation so that they may mature fully. Under a high-power (40x) microscope, the ascospores, aleroconidia, endoconidia, and perithecia from Raichur isolates the pure culture were examined to establish the fungus' identity. According to Sharma *et al.* [4], pathogen studies have been identified.

***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 the procedure for poisoned food, 20 ml of oat meal agar medium was first combined with fungicides and poured into petri

dishes with a 90 mm diameter, while the control treatment was kept going without the addition of fungicides. Five-millimeter *C. fimbriata* discs were positioned in the center of the plate after solidification. Plates were incubated at 26 ± 2 ºC and each experiment was repeated four times. In the control treatment, measurements of colony diameter were collected once the test fungal growth had reached the petriplate's edge. Later, the formula was used to compute the percent inhibition of growth (Vincent) [8].

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

Where,
I = Percent inhibition,
C = Radial growth of fungus in control
T = Radial growth of fungus in treatment.

Results and Discussion

Hexaconazole 4% + zineb 68%, carbendazim 12% + mancozeb 63%, trifloxystrobin 25% + tebuconazole 50%, and captan 70% + hexaconazole 5% were the most effective of the seven combi-fungicides evaluated against *C. fimbriata* (Table 1). Recorded maximum inhibition of 100% mycelial growth at all concentrations (0.10%, 0.20%, and 0.30%), and moderate inhibition was seen in tricyclazole 18% + mancozeb 62% (86.03%) at 0.10% concentration, 95.52% inhibition at 0.20% concentration, and 100% inhibition at 0.30% concentration. At concentrations of 0.10%, 0.20%, and 0.30%, carboxin 37.5% + thiram 37% showed 88.29% inhibition, 94.48% inhibition, and 100% inhibition, respectively. Cymoxanil 8% + mancozeb 64% showed the least amount of inhibition (89.96%) (Table 1 and Plate 1).

Hexaconazole 4% + zineb 68%, carbendazim 12% + mancozeb 63%, trifloxystrobin 25% + tebuconazole 50%, and captan 70% + hexaconazole 5% were among the combi-fungicides evaluated *in vitro*. maximal (100%) suppression of mycelial growth was seen at all doses (0.10, 0.20, and 0.30%) and while carboxin 37.5% + thiram 37% recorded 88.29% inhibition at 0.10% concentration, 94.48% inhibition at 0.20% concentration, and 100% inhibition at 0.30% concentration (Fig. 1). The earlier researchers Vijaya *et al*. [7], Somasekhara [5], Khosla [2], Sonyal *et al*. [6], and Chaudhari *et al*. [1] provide support for these conclusions.

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

Sl. No.	Fungicide	Percent inhibition at different concentrations (%)			
		0.10	0.20	0.30	Mean
1	Hexaconazole 4% + Zineb 68%	100.0 (90.0)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)*
2	Tricyclazole 18% + Mancozeb 62%	86.03 (68.05)	95.52 (77.78)	100.00 (90.00)	93.85 (78.61)
3	Carbendazim 12% + Mancozeb 63%	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
4	Carboxin 37.5% + Thiram 37%	88.29 (69.99)	94.48 (76.41)	100.00 (90.00)	94.25 (78.8)
5	Trifloxystrobin 25% + Tebuconazole 50%	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
6	Captan 70%+ Hexaconazole 5%	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)	100.00 (90.00)
7	Cymoxanil 8% + Mancozeb 64%	78.26 (62.21)	91.63 (73.18)	100.00 (90.00)	89.96 (75.13)
	Mean	81.6 (70.03)	85.20 (73.42)	87.50 (78.75)	84.76 (74.06)

	S.Em.±	CD at 1 %
Fungicides (F)	0.51	1.93
Concentration (C)	0.33	1.26
F x C	0.88	3.35

* Figures in parenthesis arc sine transformed value

Plate 1: *In vitro* evaluation of different combi-fungicides against *C. fimbriata*

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

Conclusion

Hexaconazole + zineb, carbendazim + mancozeb, trifloxystrobin + tebuconazole and captan + hexaconazole were found to be the most efficient combi-fungicides against *C. fimbriata*.

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