In vitro evaluation of fungicides against Fusarium oxysporum f. sp. ciceri

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DOI: https://doi.org/10.22271/chemi.2021.v9.i1i.11302

Abstract
Chickpea (Cicer arietinum L.) is one of the most important pulse crop grown all over India. Chickpea wilt caused by Fusarium oxysporum f. sp. ciceri is one of the major disease on chickpea in Northern Karnataka, which is soil and seed borne. Heavy inoculum in the soil and favorable environment condition results in the death of infected plant and therefore total yield loss. Experiment was conducted for selection of superior fungicides for the management of chickpea wilt disease, three combi product fungicides, three systemic fungicides and three non-systemic fungicides were evaluated against Fusarium oxysporum f. sp. ciceri in in-vitro condition with two concentrations each. Among the non-systemic fungicides copper oxychloride was effective in inhibiting the mycelial growth with 82.97% and 86.02% at 2000 and 3000 ppm concentration. Among the systemic fungicides carbendazim exhibited 79.81% and 82.03% inhibition of mycelial growth at 1000 and 2000 ppm concentration. Among the combi product fungicides Carbendazim12%+ Mancozeb 63% exhibited 100% inhibition of mycelial growth at 1000 and 2000 ppm concentration.

Keywords: Chickpea wilt, fusarium oxysporum f. sp. ciceri, fungicides

Introduction
Chickpea is (Cicer arietinum L.) is one of the most important and oldest pulse crop after beans and peas. Chickpea seeds contain an average of 23 per cent protein, 38-59 per cent carbohydrate, 4.8-5.5 per cent oil, 47 per cent starch, 5 per cent fat, 6 per cent crude fibre, 6 per cent soluble sugar and 3 per cent ash, minerals such as calcium (202 mg), phosphorous (312 mg), iron (10.2 mg), vitamin C (3.0 mg), calorific value (360 cal), small amounts of B complex, fibre (3.9 g) and moisture (9.8 g). There are two main commercial types of chickpea. The Desi type with smaller and darker coloured seeds which may vary from yellow to black and the Kabuli type with large, smooth and light coloured seeds (Singh, 1985) [10]. Chickpea crop is attacked by 172 pathogens (67 fungi, 22 viruses, 3 bacteria, 80 nematodes and phytoplasma) across the world (Nene et al., 1996) [8]. Among all, only a few of them have the potential to devastate the crops. Some of the serious diseases in order of their importance are wilt, dry root rot, collar rot, colletotrichum blight, alternaria blight, rust and ascochyta blight caused by Fusarium oxysporum f. sp. ciceri, Macrophomina phaseolina, Sclerotium rolfsii, Colletotrichum dematium, Alternaria alternata, Uromyces ciceris-arietini and Ascochyta rabiei respectively (Nene et al., 1984) [8]. Losses of chickpea from Fusarium wilt have been reported to vary from 10 to 15 per cent (Jalali and Chand., 1991; Trapero-Casas and Jimenez-Diaz., 1985) [4, 11] but losses of up to 70 per cent have been reported in some years in Northern India and Pakistan (Grewal and Pal., 1970) [2]. As a facultative saprophyte, Fusarium oxysporum f. sp. ciceri can survive in soil and on crop residues as chlamydospores for upto six years. The pathogen is also seed-borne and may therefore be spread by means of infected seed (Haware et al., 1978) [3]. Fusarium oxysporum f. sp. ciceri is considered to be a major threat to chickpea production in India, Iran, Pakistan, Nepal, Burma, Spain and Tunisia (Jalali and Chand., 1991) [4]. In the light of present day, constraints in plant disease management practices especially those on the use of botanicals and bioagents is increasingly occupying the minds of scientists all over the world as they are eco-friendly and cost effective. These antagonistic organisms act on the pathogen by different mechanisms viz., competition, lysis, antibiosis, siderophore production and hyperparasitism (Vidyasekaran, 1999). Formulations of antagonistic organisms are available at cheaper rate and these organisms once introduced into the soil survive for a longer period.
The pathogen is mainly soil borne thus seed treatment by fungicides is considered the easiest and most economical way for management of the disease. Therefore the present study was carried out to evaluate the fungicides against Fusarium wilt of chickpea under in vitro.

### Material and Methods

**Isolation of Fusarium oxysporum f.sp. ciceri**

Fusarium oxysporum f. sp. ciceri was isolated from infected chickpea plants from different districts of Northern Karnataka, India. The Fusarium oxysporum f. sp. ciceri was identified, purified and preserved in PDA medium and confirmation of F. oxysporum f. sp. ciceri by Koch’s postulation and based on the morphological characters described by Booth (1971) [1].

**In vitro evaluation of fungicides against Fusarium oxysporum f. sp. ciceri**

Required quantity of individual fungicide was added separately into sterilized molten and cooled potato dextrose agar so as to get the desired concentration of the fungicides through poisoned food technique (Nene and Thapliyal, 1973) [6]. Later, 20 ml of the poisoned petriplate. Mycellial disc of five mm size from actively growing zone of seven days old culture was cut by a sterile corn borer and one such disc was placed at the centre of each agar plate. Control treatment was maintained without adding any fungicide to the medium. Three replications were maintained for combi product and systemic fungicides and seven replications were maintained for non-systemic fungicides. Then such plates were incubated at room temperature and radial growth was measured when fungus attained maximum growth in control plates. Per cent inhibition of mycelial growth over control was calculated by using the formula given by Vincent (1947) [12].

\[
I = \frac{(C - T)}{C} \times 100
\]

Where

- **I** = Per cent inhibition
- **C** = Radial growth in control
- **T** = Radial growth in treatment

### Table 1: Details of fungicides used in the experiment.

| Sl. No. | Common name               | Chemical name                                                                 | Trade name  |
|---------|---------------------------|-------------------------------------------------------------------------------|-------------|
| 1       | Captana                   | N-trichloromethyl mercapta-4-cyclohexene-1,2-dis-carboximide N-trichloromethyl thiotetrahydrothalamide | Captop 50 WP |
| 2       | Wettable Sulphur          |                                                                                | Wettasul 80%WP |
| 3       | Copper oxychloride        | Copper chloride oxide, hydrate                                               | Blixt 50 WP |

Contact fungicides were evaluated at 0.2 and 0.30 per cent

### Table 2: Chemical name

| Sl. No. | Common name | Chemical name                                                                 | Trade name  |
|---------|-------------|-------------------------------------------------------------------------------|-------------|
| 1       | Hexaconazole| (RS) -2- (2,4-dichlorophenyl) -1- (14-1,2,4-triazole-1 YL) hexane-2-1        | Contaf 5% EC |
| 2       | Propiconazole| 1- (2,4 di chlorophenyl) –4-ropyl- 1,3-dioxolan-2-methyl –H-1,4- triazole | Tilt 25EC   |
| 3       | Carbendazim | 2- (methoxy-carbomyl) - benzimidazole                                         | Bavistin 50% WP |

Systemic fungicides were evaluated at 0.1 and 0.2 per cent

### Table 3: Show the chemical name

| Sl. No. | Common name               | Chemical name                                                                 | Trade name  |
|---------|---------------------------|-------------------------------------------------------------------------------|-------------|
| 1       | Carbendazim 12% + Mancozeb 63% WP | Methyl benzimidazole carbonate + Manganese zinc ethylene BIS dithiocarbamate + zinc | SAAF 75% WP |
| 2       | Captan70% + Hexaconazole 5% WP | N-trichloromethyl mercapta-4-cyclohexene-1,2-dis-carboximide N-trichloromethyl thiotetrahydrothalamide + RS –2- (2,4-dichlorophenyl) –1- (14-1,2,4-triazole-1 yl) hexane-2-1 | Taquat 75% WP |
| 3       | Carboxin 37.5% + Thiram 37.5% | 3-(3-5-dichlorophenyl) –N- (1- methylethyl) –2-4-dioxo-1- lamadazolidine carboximide + tetramethyl thiram disulphide | Vitavax power 75% WP |

Combi products were evaluated at 0.1 and 0.2 per cent

### Results and Discussion

Three contact fungicides were tested against F. oxysporum f. sp. ciceri, among which copper oxychloride gave the best result by maximum inhibition of 84.49% which was significantly superior to all other fungicides and the least inhibition of the mycelial growth was in wettable sulphur (51.50%). Copper oxychloride gave best result at 0.30 per cent concentration in inhibiting the growth of pathogen by (86.02%) and least inhibition was observed in the wettable sulphur @ 0.30 per cent (53.18%) (Table 1). Three systemic fungicides were evaluated against F. oxysporum f. sp. ciceri, among which carbendazim was found best with 80.92 per cent inhibition at 0.1 and 0.2 per cent concentration and significantly superior to all other treatments in inhibiting growth of Fusarium oxysporum f. sp. ciceri. This was followed by Hexaconazole with 70.46 per cent inhibition. The least inhibition of mycelial growth was observed in propiconazole with 63.82 per cent (Table 1, 5.). Among the different concentrations, carbendazim was effective @ 0.2 per cent with 82.03 per cent inhibition and significantly superior to all fungicides the least inhibition was found in the propiconazole at @ 0.2 per cent with inhibition of 65.43 per cent. Among three combi products evaluated, carbendazim 12% + mancozeb 63% (SAAF) was found to be most effective and significantly superior to all other fungicides, which inhibited cent per cent growth of F. oxysporum f. sp. ciceri. This was followed by carboxin 37.5% + thiram 37.5% (Vitavax power 75% WP) with 80.82 per cent inhibition and the least inhibition of mycelial growth (76.01%) was observed captan 70% + hexaconazole 5% WP (Taquat).
Among different concentrations tested carbendazim 12% + mancozeb 63% (SAAF) @ 0.1 per cent and 0.2 per cent inhibited cent per cent which was significantly superior all other treatments and least inhibition (74.81% and 77.22%) was recorded in captan 70% + hexaconazole 5% WP (Taquat) @ 0.1 and 0.2 per cent respectively. (Table 2).

Among all three groups of fungicides, systemic fungicides and combi products were found to be more effective than non-systemic fungicides. Among the non-systemic fungicides tested against Fusarium oxysporum f. sp. ciceri, the average highest per cent inhibition was observed in copper oxychloride at 0.30 per cent with (86.02) per cent. Similar results were reported by Ravichandran (2015) [9]. Among systemic fungicides evaluated against Fusarium oxysporum f. sp. ciceri, the average highest per cent inhibition was observed in carbendazim (80.92) per cent, which was significantly superior to all other fungicides. The average lowest per cent inhibition was observed in propiconazole (63.82) per cent. Among the different concentrations tested, significantly highest mean inhibition was recorded in carbendazim at 0.20 per cent with 82.03 per cent. (Table 2). Similar results were reported by Ravichandran (2015) [9] and Mahmood et al. (2015) [5]. Among combi product fungicides evaluated against Fusarium oxysporum f. sp. ciceri, the average cent per cent inhibition was observed in carbendazim 12% + mancozeb 63%, which was significantly superior to all other combi product fungicides and the average lowest per cent inhibition was observed in captan 70% + hexaconazole 5% WP 76.01 per cent. Among the different concentrations tested, significantly highest mean cent per cent inhibition was recorded in carbendazim 12% + mancozeb 63% at 0.20 per cent. (Table 2). Similar results were reported by Ravichandran (2015) [9].

### Table 4: In vitro evaluation of non-systemic fungicides against Fusarium oxysporum f. sp. Cicero

| Sl. No. | Fungicides          | Trade name                  | Inhibition of mycelial growth (%) | Mean |
|---------|---------------------|-----------------------------|-----------------------------------|------|
|         |                     |                             | 0.20%          | 0.30%          |      |
| 1       | Wettlesul 80% WP    | Wetsul 80% WP               | 49.83 (44.92)* | 53.18 (46.84)  | 51.50 (45.88) |
| 2       | Captan 50% WP       | Captaf 50% WP               | 60.75 (51.23)  | 65.03 (53.77)  | 62.89 (52.49) |
| 3       | Copper oxychloride  | Blitox-50% WP               | 82.97 (65.66)  | 86.02 (68.07)  | 84.49 (66.84) |
|         | Mean                |                             | 64.51 (53.46)  | 68.07 (55.62)  | 66.29 (54.53) |
|         | S.Em. ± C.D. at 1%  |                             | 0.40            | 1.55            |      |

*Arcsine transformed values

### Table 5: In vitro evaluation of systemic and combi products fungicides against Fusarium oxysporum f. sp. Cicero

| Sl. No. | Fungicides                  | Trade name | Inhibition of mycelial Growth (%) | Mean |
|---------|-----------------------------|------------|-----------------------------------|------|
|         |                             |            | 0.1                     | 0.2             |
| 1       | Carbendazim 12% + Mancozeb 63% | Saaf       | 79.81 (63.30) | 82.03 (64.92) | 80.92 (64.10) |
| 2       | Hexaconazole 5% EC          | Contaf 5%  | 68.33 (55.75) | 72.59 (58.43) | 70.46 (57.08) |
| 3       | Propiconazole 25% EC        | Tilt 25% EC| 62.21 (52.07) | 65.43 (53.99) | 63.82 (53.02) |
|         | Mean                        |            | 84.69 (67.00) | 86.54 (68.51) | 85.61 (67.74) |

*Arcsine transformed value
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