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# Evaluation of bio-efficacy and phyto-toxicity of flint-pro (Trifloxystrobin 3.5% + Propineb 61.3% WG) against anthracnose disease and leaf spot disease of chilli

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**Abstract**
Chilli (*Capsicum annum*) is an important cash crop belonging to the family Solanceace. It is mainly used for pungency in cooking items or can be eaten as raw. In powder form, it can be used as a bioinsecticides. The crop undergo various biotic and abiotic stresses. Among the biotic stresses, anthracnose and leaf spot caused by *Colletotrichum* spp. and *Alternaria alternate* are the two serious problem causing a heavy loss in yield and productivity. Various biocontrol or botanical are used to control chilli anthracnose and leaf spot disease but with little or no effect on it. A number of chemicals are also recommended to curb the problem but with a little affect in the long run and the chemical become obsolete. Therefore, present study was carried out to evaluate the bio-efficacy and phyto-toxicity of fin-pro (Trifloxystrobin 3.5% + Propineb 61.3% WG) on Chilli against anthracnose disease and leaf spot disease in field condition. Eight treatment combinations were tested against anthracnose and leaf spot of chilli in two consecutive years. Pooled data showed that out of all the treatments T8 i.e, Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) was found best in suppressing both the diseases with low phytotoxicity. This treatment may be recommended for management of both the diseases in field condition.

**Keywords:** Evaluation, bio-efficacy, phyto-toxicity, anthracnose

## Introduction
Chilli (*Capsicum annuum* L.) is an imperative spice, as well as vegetable in every household of India. It has an important role in our daily diet and considered as an important vegetable as well as commercial crop. Chilies are having good nutritious value and can be used as multipurpose throughout the world for its aroma, pungency and medicinal value. Chilli also has uncountable benefits to human health. Fresh green chilli fruits contain more Vitamin C than found in citrus fruits, while red chilli fruits have more Vitamin A content than as found in carrots. The active component of the spice, Capsaicin possesses the antioxidant, anti-mutagenic, anti-carcinogenic and immune suppressive activities having ability to inhibit bacterial growth and platelet aggregation. In India chilli has been grown in an area of 287.05 million Ha with production of 3406.03 metric tonne (Source: Horticulture Statistics Division, Department of Agriculture, Corporation & Farmers Welfare 2016-17). Assam occupies an area of 21.31 million Ha with a production of 19.12 metric tonne. (Source: Horticultural statistics at a glance, 2017).

Anthracnose disease is one of the major economic constraints to chilli production worldwide, especially in tropical and subtropical regions, reducing marketable yield from 10% to 80% of the crop production in some developing countries, particularly in Thailand [1]. Anthracnose is mainly a problem on mature fruits, causing severe losses due to both pre- and post-harvest fruit decay [2, 3].

Little information is known concerning the interactions of the species associated with the chilli anthracnose although several *Colletotrichum* species have been reported as causal agents of chilli anthracnose disease worldwide. Four species of Colletotrichum; *C. capsici*, *C. gloeosporioides*, *C. acutatum* and *C. coccodes* have been reported as causal agents of pepper anthracnose in many countries. The major species are *C. capsici* and *C. gloeosporioides* [4]. Typical anthracnose symptoms on chilli fruit include sunken necrotic tissues, with concentric rings of acervuli. Fruits showing blemishes have reduced marketability [5]. Although the management and control of anthracnose disease are still being extensively researched,

commercial cultivars of *Capsicum annuum* that are resistant to the pathogens that cause chilli anthracnose have not yet been developed. Many studies have concluded that disease management practices are often inadequate to eliminate the diseases. Breeding to develop the long-lasting resistant varieties has also not been successful due to involvement of multiple *Colletotrichum* species in anthracnose infection. In India, a calculated loss of 10-54 per cent has been reported in yield of the crop due to the anthracnose disease [6]. The loss is high owing to the post and pre-harvest involvement of the pathogen causing a loss of 10-80 per cent of the marketable yield of chilli fruits [7].

The leaf spot of chilli caused by *Alternaria alternata* (Fr) Keissler is becoming a limiting factor and posing a major problem in chilli production and considered as destructive disease in chilli growing areas of India. The pathogen is seed borne and reduce the seed germination and yield loss up to 30-60 per cent. The pathogen has been reported to cause seed seeding, leaf and fruit diseases as well [8, 9, 10]. Post harvest decay of fruits and seeds has also been recorded due to this pathogen [11]. The current study evaluates the bio-efficacy and phyto-toxicity of flint-pro (Trifloxystrobin 3.5% + Propineb 61.3% WG) against anthracnose disease and leaf spot disease of chilli.

## Materials and Methods

The study was carried out during the session 2016-17 and 2017-18 at the Department of Plant Pathology, Assam Agricultural University, Jorhat. Field experiments was conducted in the instruction cum research (ICR) farm, AAU during 2016-17 and 2017-18 in *rabi* season. The materials used and the methods applied during the experiments are described below.

### Cultivation practices

Two susceptible variety was used namely Krishna Jolokia and Kancha for the present study. Standard crop cultivation practices was adopted as per the package of practice for Assam. Crop was grown in Sandy loam soil and fertilized at 120:80:80 (NPK) Kg/ha. Eight treatment combinations as mentioned in the table 1 were tested each replicated for four times. All the treatments were sprayed for five times at 10 days intervals with Knapsack sprayer as per the formulated treatment combinations.

**Table 1:** Treatment combination used for to study the effect on anthracnose and leaf spot of chilli

| Tr. No. | Treatments | Dosage/Ha | | Method of Application |
|---|---|---|---|---|
| | | a.i.(g) | a.i.(g) | |
| 1 | Untreated Control | - | - | - |
| 2 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 52.5+919.5 | 1500 | First Spray at the onset of disease. Subsequent spays at 10 days interval. The product was used for foliar spray as well as soil drenching with a recommended dose of 2ml/lit (as foliar spray) and 3ml/lit (as soil drenching). Water Volume @ 650lit/ha |
| 3 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 61.2+1072.7 | 1750 | |
| 4 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 70+1226 | 2000 | |
| 5 | Trifloxystrobin 50% WG | 70 | 140 | |
| 6 | Propineb 70% WP | 1750 | 2500 | |
| 7 | Captan 75% WP | 1500 | 2000 | |
| 8 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG)* | 140+2452 | 4000 | |

**Observations on anthracnose disease caused by *Colletotrichum capsici:*** Per cent disease severity/index as per standard method is recorded from 5 randomly tagged plants at pre-treatment and at 5 &10 days after each spray. The percent disease severity recorded by using the 0-5 scale as proposed by Jeyalakshmi and Seetharaman [12] and disease index was calculated by the following formula. Where 0- no disease, 1= up to 5%, 2= 5 to 10%, 3= 10 to 25%, 4= 25 to 50% and 5= above 50%.

$$\text{The anthracnose percent/disease index} = \left[\frac{\text{Sum of disease ratings}}{\text{Total no. of ratings (Fruits) x maximum disease rating}}\right] \times 100$$

**Observations on leaf spot disease caused by *Alternaria solani*:** Per cent disease severity/index was calculated as per 0-5 scale arbitrary scale (where, 0 = no visible symptoms of disease, 1 = lesions covering 1- 20% area of infected leaves, 2 = lesions covering 21-40% area of the infected leaves, 3 = lesions covering 41-60% area of infected leaves, 4 = lesions covering 61-80% area of infected leaves, 5= lesions covering 81- 100% area of infected leaves). Data were recorded from 5 randomly tagged plants at pre-treatment and at 5 and 10 days after each spray.

### Phytotoxicity

Data on phytotoxicity were recorded as per the Phytotoxicity Rating Scale (PRS). Ratings were recorded individually for yellowing, stunting, necrosis, epinasty and hyponasty.

**Table 2:** Phytotoxicity Rating Scale (PRS)

| Crop response/Crop injury | Ratings |
|---|---|
| 0-00 | 0 |
| 1-10% | 1 |
| 11-20% | 2 |
| 21-30% | 3 |
| 31-40% | 4 |
| 41-50% | 5 |
| 51-60% | 6 |
| | |
| 61-70% | 7 |
| 71-80% | 8 |
| 81-90% | 9 |
| 91-100% | 10 |

**Yield**

Total chilli yield from all the pickings per plot was recorded and converted to yield/ha.

**Statistical analysis**

Randomized block design were followed for pot test and field experiment respectively for the statistical analysis of the data. The data collected were subjected to statistical analysis by Fisher's method of analysis of variance. Significance of variance among the data was analysed by calculating the "F" value and comparing it with the tabulated value of "F" at 5% level of probability.

The treatment means were compared among themselves by calculating critical difference (CD) as followed:

C.D. at 5% = S. Ed ×'t' 5% (at error d.f.)

The standard error of differences (S. Ed) was calculated by using the following formula:

$$SEd. = \sqrt{\frac{2 \times \text{Error Mean Square}}{\text{Number of replication for each treatment}}}$$

Where, SEd = Standard error of difference

't' 5% = "t" for error d.f. at 5% level of probability

The significance and non-significance of the treatments at 5% level of probability were calculated out by multiplying the SEd., with appropriate tabulated value for error degrees of freedom.

Analysis of variance (ANOVA) tests were performed on data to test for significant ($P<$ 0.05) differences between fungicides. Least significant difference (LSD) test was used to compare means of treatments. Percentage values were analyzed after arcsine transformation of the raw data.

**Results and Discussion**

**Efficacy on Anthracnose disease**

After the final spray (i.e. after 10 days of spraying) treatment $T_8$ Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 140g+2452g) was the best followed by $T_3$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 61.2g+1072.7g) and $T_2$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 52.5g+919.5g) respectively. $T_8$ reduced the PDI from 20.37% to 3.87%, whereas $T_3$ and $T_2$ resulted in reduction of the PDI from 20.75% to 4.62% and 5.25% respectively. $T_2$ and $T_3$ were statistically at par in their efficacy. $T_6$ (Propineb 70% WP) and $T_7$ (Captan 75Wp) were also statistically at par in reducing the PDI. Both the treatments reduced the PDI from 20.00% to 12.37% and 20.25% to 11.50% respectively. Least reduction of PDI was recorded in Trifloxystrobin 50% WG (Table 3). Hegde *et al.* [13] reported that fungicides like Hexaconazole (0.1%), Propiconazole (0.1%) and Triademefon (0.1%) were effective against the fruit rot pathogen (*C. capsici*) of chilli that inhibited mycelial growth significantly. Another study conducted by Gopinath *et al.* [14] found that Propiconazole exhibited the highest level of mycelial growth inhibition, sporulation and spore germination at concentrations as low as 0.1 $\mu$g/ml. Carbendazim and Thiophanatemethyl found to be highly effective against *Colletotrichum capsici* @ 0.1% [15, 16]. Singh *et al.* [17] observed that maximum reduction in fungal growth was obtained for carbendazim and Triademefon followed by propiconazole. It was reported that the anthracnose causing pathogen, *C. capsici* were highly sensitive to Chlorothalonil followed by Propiconazole, Aliette and Mancozeb respectively in chilli crop [18, 19]. In a study conducted by Gopinath *et al.* [14] observed the highest inhibition of enzyme production in Propiconazole, Difenoconazole and Carbendazim.

**Efficacy on Leaf spot disease**

After the final spray (i.e. after 10 days of spraying) treatment, best result in terms of PDI was recorded in $T_8$ Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 140g+2452g), followed by $T_4$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 70g+1226g), $T_3$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 61.2g+1072.7g) and $T_2$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 52.5g+919.5g). $T_8$ reduces the PDI from 10.58% to 4.95%, whereas $T_4$ reduces the PDI from 10.41% to 6.55%. The efficacy recorded for $T_3$ and $T_2$ were found to be statistically at par with each other. Reduction of the PDI recorded in $T_3$ was from 10.42% to 7.18% whereas in $T_2$ reduction of PDI was from 10.50% to 7.60%. The least efficacy was found in $T_6$ (Propineb 70% WP) followed by $T_7$ (Captan 75Wp). The treatments reduced the PDI from 10.60% to 9.08% and 10.62% to 8.30% respectively (Table 3).

Tu and Somasekhara [20] reported complete mycelial growth inhibition of *Alternaria brassicicola* by the fungicides Propiconazole and Tebuconazole. In an another study, Singh and Singh [21] reported that the fungicide hexaconazole showed 100% inhibition of *Alternaria alternata*, that causes alternaria blight of tomato. It was also reported that tebuconazole and hexaconazole completely inhibited the mycelial growth of *Alternaria alternata* in chilli [22]. Kiran *et al.* [23] tested the efficacy of strobilurin fungicides against *Alternaria dauci* and revealed that the percentage of inhibition was 13.33% with azoxystrobin and 14.44% with Trifloxystrobin.

**Table 3:** Percent Diseases incidence of anthracnose and leaf spot disease of chilli

| Treatments | % disease incidence at Pre treatment | | % disease incidence at 5 days after spray | | % disease incidence at 10 days after spray | |
|---|---|---|---|---|---|---|
| | Anthracnose | Leaf spot | Anthracnose | Leaf spot | Anthracnose | Leaf spot |
| T1 | 20.37$^{a}$ | 10.35$^{a}$ | 23.00$^{e}$ | 12.03$^{f}$ | 26.12$^{f}$ | 15.38$^{g}$ |
| T2 | 20.75$^{a}$ | 10.50$^{a}$ | 16.25$^{ab}$ | 9.35$^{cd}$ | 5.25$^{b}$ | 7.60$^{cd}$ |
| T3 | 20.75$^{a}$ | 10.42$^{a}$ | 16.50$^{abc}$ | 9.03$^{bc}$ | 4.62$^{ab}$ | 7.18$^{cd}$ |
| T4 | 20.75$^{a}$ | 10.41$^{a}$ | 16.87$^{abcd}$ | 8.83$^{bc}$ | 10.75$^{c}$ | 6.55$^{b}$ |
| T5 | 20.37$^{a}$ | 10.64$^{a}$ | 18.37$^{d}$ | 9.93$^{e}$ | 13.87$^{e}$ | 8.13$^{de}$ |
| T6 | 20.00$^{a}$ | 10.60$^{a}$ | 18.12$^{cd}$ | 9.58$^{de}$ | 12.37$^{d}$ | 9.08$^{f}$ |
| T7 | 20.25$^{a}$ | 10.62$^{a}$ | 17.87$^{bcd}$ | 9.00$^{bc}$ | 11.50$^{cd}$ | 8.30$^{e}$ |
| T8 | 20.37$^{a}$ | 10.58$^{a}$ | 15.87$^{a}$ | 8.28$^{a}$ | 3.87$^{a}$ | 4.95$^{a}$ |
| SEM | 0.86 | 0.22 | 0.58 | 0.21 | 0.61 | 0.27 |
| CD | 1.78 | 0.45 | 1.99 | 0.44 | 1.26 | 0.56 |

*Data are pooled of two seasons experiments

**Phytotoxicity**

Yellowing and partial epinasty of the leaves were observed in the $T_4$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) a.i. 70g+1226 g) and $T_8$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) a.i. 140g+2452 g). PRS rating was found more in $T_8$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) a.i. 140g+2452 g). This may be due to higher dose of the active ingredients. PRS rating recorded for $T_8$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) a.i. 140g+2452 g) is 2, whereas for the $T_4$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) a.i. 70g+1226 g) it was found to be 1. (Table 4)

**Table 4:** Phyto-toxicity of Flint-Pro (Trifloxystrobin 3.5% + Propineb 61.3% WG) on Chilli against anthracnose disease and leaf spot disease

| Tr. No. | Treatment | Dosage/ha | | Days after 1st Spray | | | | |
|---|---|---|---|---|---|---|---|---|
| | | a.i. (g) | Formulations (g) | 1 | 3 | 5 | 7 | 10 |
| 1 | Untreated Control | - | - | 0 | 0 | 0 | 0 | 0 |
| 4 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 70+1226 | 2000 | 1 | 1 | 1 | 1 | 1 |
| 8 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 140+2452 | 4000 | 2 | 2 | 2 | 2 | 2 |

**Yield**

Highest yield was observed in T8 (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 140g+2452g), followed by $T_3$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 61.2g+1072.7g) and $T_2$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 52.5g+919.5g). The increase in yield as compared to untreated control was mainly due to the efficacy of the fungicide treatments on anthracnose and leaf spot infection (Table 5)

**Table 5:** Yield data due to the application of Flint-Pro (Trifloxystrobin 3.5% + Propineb 61.3% WG) on Chilli against anthracnose disease and leaf spot disease

| Tr. No. | Treatments | Dosage/Ha | | Yield/ha (Kg/ha) |
|---|---|---|---|---|
| | | a.i.(g) | a.i.(g) | |
| 1 | Untreated Control | - | - | 189.28 |
| 2 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 52.5+919.5 | 1500 | 434.00 |
| 3 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 61.2+1072.7 | 1750 | 470.00 |
| 4 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG) | 70+1226 | 2000 | 352.00 |
| 5 | Trifloxystrobin 50% WG | 70 | 140 | 256.06 |
| 6 | Propineb 70% WP | 1750 | 2500 | 240.64 |
| 7 | Captan 75% WP | 1500 | 2000 | 226.00 |
| 8 | Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG)* | 140+2452 | 4000 | 744.00 |

**Conclusion**

From this experiment we can conclude that treatment $T_8$ Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 140g+2452g) as the best treatment in terms of reduction both the diseases and with increase in yield and lowest phytotoxicity effect. This was followed by $T_3$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 61.2g+1072.7g) and $T_2$ (Flint Pro (Tri-floxystrobin 3.5 + Propineb 61.3% WG, a.i. 52.5g+919.5g).

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