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EFFECT OF TAEGRO AS BIO-FUNGICIDE ON YIELD AND CONTROL OF EARLY AND LATE BLIGHT DISEASES IN TOMATO (*Solanum lycopersicum* L.)

*Manukinda Adinarayana, Narasegowda, N. C., Premalatha, Y., Anjanappa, M., Venkata Subbaiah Yadav, B. Devappa, D.
*Department of Vegetable Science, COH, UHS Campus, GKVK, Bangalore - 560 065, Karnataka, India.
*Corresponding author email: adihorti007@gmail.com

ABSTRACT
An experiment to study the effect of taegro, a bio-fungicide, on yield and control of early and late blight diseases in tomato (*Solanum lycopersicum* L.) cv. NS -501 was conducted at the Department of Horticulture, Gandhi Krishi Vignana Kendra, University of Agricultural Science, Bangalore during *rabi* 2013-2014. The study included: seed treatment with taegro (4g/kg), seedling dip with taegro solution (4g/l), both seed treatment with taegro (4g/kg) and soil drenching (250g/ha), both seedling dip with taegro (4g/l) and soil drenching (500g/ha), both soil drenching and foliar spray with taegro at 250g/ha and both soil drenching and foliar spray with taegro at 500g/ha, mancozeb @ 2g/l as standard fungicide and untreated control. Among all tested combination, the treatment containing combination of bio-fungicide (taegro) by both soil drenching and foliar spray @ 500 g per hectare has proved to be the most appropriate and economically viable treatment in enhancing yield and disease control. The next best treatment was soil drenching and foliar spray of taegro @ 250 g per hectare. However, the standard chemical mancozeb (2g/l) recorded highest number of fruits per plant (110.07), yield per plant (6.42 kg), yield per plot (179.42 kg), yield per hectare (117.33 t) and least diseases incidence of *Alternaria solani* of 8.97 and 15.79 PDI at 65 DAT and 90 DAT, respectively and *Phytophthora infestans* of 5.13 and 14.69 PDI at 65 DAT and 90 DAT, respectively.

KEYWORDS*:* Bio-fungicide, Taegro, Mancozeb, *Alternaria solani, Phytophthora infestans.*

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is grown in an area of 0.879 million hectare with a production of 18.22 million tonnes and the productivity being 20.72 tonnes per hectare. The leading tomato growing states are Andhra Pradesh, Karnataka, Orissa, West Bengal, Maharashtra, Haryana, Uttar Pradesh, Punjab and Bihar. In Karnataka, it occupies an area of 0.57 lakh hectares with a production and productivity 19.16 lakh tonnes and 33.15 tonnes/ha respectively[1]. The leading tomato growing districts in the state are Belgaum, Dharwad, Kolar, Bengaluru and Bellary. Tomato plant is attacked from many serious diseases under greenhouse and field conditions. Several important diseases of tomato reduce crop yield and the most devastating plant pathogens are fungi and oomycetes [2]. For example, the early blight, caused by *Alternaria solani* (Ellis & Martin) Sorauer, [3] and late blight, incited by *Phytophthora infestans* (Mont.) deBary[4] are economically important diseases of tomato worldwide including India causing crop losses up to 100%[3]. Since commercial cultivars do not have sufficient resistance to leaf blights, cultural practices and fungicides applied at 5–7 days intervals form the basis for leaf blight management programs[5]. However, development of fungicide resistance, accumulation of residues in fruits, reduction of beneficial phylloplane and soil microbes and environmental pollution are associated problems[6]. Considering the seriousness of the problem, the present investigation was carried out. The hazardous effects of chemicals used in plant disease management have diverted plant pathologists to find out the alternative techniques of plant disease control which may cause little or no adverse effect on environment. Notable success of disease management through the use of antagonistic bio-agents in the laboratory, glass house and field has been achieved during past several years. On the basis of this information, there is possibility of development of biological control for plant diseases. Now a day, the commercial formulation of some of the bio-control agents has already become available in the market. In the present study, attempts have been made to identify antagonistic bio-agents against early and late blight diseases in field condition.

Plant Growth Promoting Rhizobacterium (PGPR) such as *Bacillus subtilis* (Ehrenberg) Cohn is used in a wide range of crop plants as biocontrol agent for management of different pathogens[7]. Induced systemic defense responses in plants have been reported as one of the mechanisms by which these organisms reduce the diseases in plants in conjunction with other mechanisms including direct antagonism, antibiosis and siderophore production. Induction of defense responses by *Bacillus* spp. is largely associated with production of pathogenesis related proteins like b-1,3-glucanase and the defense enzyme phenylalanine ammonia-lyase and oxidative enzymes like peroxidase, polyphenol oxidase and superoxide dismutase [8]. Apart from controlling diseases, these bio-control organisms also promote plant growth by production of plant growth hormones like IAA and GA_3 coupled with increased availability of nutrients [7].

Taegro is novel bio-fungicide released by the company Novozymes South Asia Pvt. Ltd. Taegro has been commercially tested on different vegetable crops in controlling diseases like tomato (late blight, bacterial wilt and bacterial spot diseases), cucumber (*Rhizoctonia*), lettuce (bottom rot and downey mildew) and pepper (powdery mildew) at the rate of 180-360 g/ha at 7-28 days interval for soil borne diseases and 7-14 days interval for foliar diseases with 1-12 number of applications during crop season. Taegro is a bio-fungicide which contains *Bacillus subtilis* var. *amyloliquefaciens* strain FZB24, which is used to suppress soil-borne diseases like *Fusarium* spp., *Rhizoctonia* spp. and *Phytophthora* spp. and also the active ingredient *Bacillus subtilis* var. *amyloliquefaciens* strain FZB24 also acts as a plant growth promoter. It also produces various antifungal agents and enzymes. Taegro involves in more than 8.5 % production of secondary metabolites (peptides, lipopeptides, polyketides and siderophores) through pathways that do not involve ribosomes. The genome contains nine giant gene clusters directing the synthesis of Lipopeptides (Surfactin, Iturins, Fengycin, Bacillibactin and Bacilysin) and Polyketides (Bacillene, Difficidin, Macrolactin). These compounds are known to suppress bacteria and fungi within the plant rhizosphere. At present agriculture / horticulture ecosystem contains high toxicity fungicides and is leading to environmental impact and also residues remaining in fruits and vegetables. The aim of this study was to determine the effects of commercially available *Bacillus amyloliquefaciens* strain FZB24 on yield and control of early and late blight diseases in field condition.

MATERIALS & METHODS

The present experiment was conducted to test the new bio-fungicide (taegro) on yield and control of early and late blight diseases of hybrid tomato cv. NS-501 was carried out under field condition. Research was undertaken at the Department of Horticulture, Gandhi Krishi Vigyan Kendra, University of Agricultural Science, Bangalore during *rabi* season of 2013-2014. The seedlings required for the experiment were raised in pro trays at Department of Horticulture, GKVK. Twenty eight days old healthy and uniform seedlings were transplanted to 15.12 m^2 experimental plots, maintaining a row spacing of 1.2 m with 0.45m between plants in a row with a population of 28 plants per plot. Before transplanting well decomposed farm yard manure at the rate of 35 tonnes per hectare was applied along with the fertilizer dose of 180:120:150 kg NPK ha^{-1}. According to the fertilizer schedule total P and K and half dose of nitrogen was applied as basal dose and the remaining half dose of nitrogen was top dressed after 30 days of transplanting and other management practices were undertaken as per the package of practices for horticultural crops given by University of Horticultural Sciences, Bagalkot. The experiment was carried out in Randomized Completely Block Design (RCBD) with eight treatments and three replications. The study included: seed treatment with taegro (4g/kg), seedling dip with taegro solution (4g/l), both seed treatment with taegro (4g/kg) and soil drenching (250g/ha), both seedling dip with taegro (4g/l) and soil drenching (500g/ha), both soil drenching and foliar spray with taegro at 250g/ha and both soil drenching and foliar spray with taegro at 500g/ha, mancozeb @ 2g/l as standard fungicide and untreated control.The observations with regard to yield and incidence of early and late blight diseases. Disease incidence was observed on five randomly chosen plants per plot at 65 and 90 days after transplanting. Five leaves were selected from different positions of each plant and the leaf area infested by each disease was measured on 0-5 scale [9] (Table 1). Each disease was identified on the basis of following symptoms and expressed as % Disease Index (PDI).

Percent disease index (PDI) was calculated as per the formula of [10] and the data was analyzed statistically.

$$\% \text{ disease index} = \frac{\text{Sum of individual ratings}}{\text{No. of leaves assessed}} \times \frac{100}{\text{Max. disease grade}}$$

TABLE 1: Percent Disease Index (PDI)

% leaf area covered	Severity scale	Disease reaction
<1	0	Immune
1-5	1	Resistant
6-20	2	Moderately resistant
21-40	3	Moderately susceptible
41-70	4	Susceptible
71-100	5	Highly susceptible

RESULTS & DISCUSSION

Effect of different methods and levels of application of taegro on number of fruits per plant, yield per plant, per plot and per hectare

The treatment T_7 (Standard chemical control by spraying mancozeb @ 2 g/l) recorded the maximum number of fruits per plant (110.07), yield per plant (6.42kg), yield per plot (179.42kg) and yield/ha (112.13t), which was *on par* with T_6, for number fruits per plant (107.40) and T_6 and T_5 for yield per plant (6.31 and 6.18kg/plant, respectively) (Table 2). The probable reason for such finding may be that, mancozeb would have affected the spore germination and mycelial development, which may have resulted in the inhibition of disease producing activity of pathogen in the plant and induced resistance in plant. This may be the reason for minimum disease intensity and maximum yield as compared to other treatments. Similar trends were reported by [11]. They also reported that mancozeb was the most effective fungicide in minimizing disease intensity against early blight of tomato. This was also supported by the findings of Chourasiya *et al.* [12] against early blight of tomato. Treatment T_7 was *on par* with T_1, T_2, T_3, T_4, T_5 and T_6 with respect to fruit yield per plot and fruit yield per hectare. The minimum number of fruits per plant (65.52), yield per plant (3.42kg), yield per plot (96.29kg) and yield per hectare (60.18t) was recorded in treatment T_8 *i.e.*, untreated control.

TABLE 2. Effect of different methods and levels of taegro as bio-fungicide on number of fruits per plant, yield per plant, per plot and per hectare of all harvests of hybrid tomato cv. NS -501

Treatments	Total of all harvests			
	Number of fruits per plant	Yield per plant (kg)	Yield per plot (kg)	Yield per hectare (t)
T_1- Seed treatment with taegro @ 4g/kg seed	98.38 ^{bc}	6.12 ^{b}	170.47 ^{b}	106.54 ^{b}
T_2- Seedling dip with taegro @ 4g/l of water for 25 minutes	94.45 ^{ab}	5.77 ^{bc}	161.26 ^{b}	100.78 ^{b}
T_3- Both seed treatment with taegro @ 4g/kg seed and soil drenching twice (*i.e.*, 15 and 30 DAT) @ 250 g/ha	89.79 ^{a}	5.30 ^{b}	147.93 ^{b}	92.45 ^{b}
T_4- Both seedling dip with taegro @ 4g/l of water and soil drenching twice (*i.e.*, 15 and 30 DAT) @ 500 g/ha	98.32 ^{bc}	6.12 ^{bc}	171.80 ^{b}	107.37 ^{b}
T_5- Both soil drenching and foliar spray with taegro @ 250g/ha on 15 and 30 DAT	102.94 ^{cd}	6.18 ^{bc}	173.56 ^{b}	108.47 ^{b}
T_6- Both soil drenching and foliar spray with taegro @ 500g/ha on 15 and 30 DAT	107.40 ^{de}	6.31 ^{c}	177.40 ^{b}	110.87 ^{b}
T_7- Standard chemical control by spraying mancozeb @ 2 g/l 10 days interval throughout crop growth period	110.07 ^{e}	6.42 ^{c}	179.42 ^{b}	112.13 ^{b}
T_8- Untreated control	65.52 ^{a}	3.42 ^{a}	96.29 ^{a}	60.18 ^{a}
F test (p=0.05)	*	*	*	*
S Em ±	2.83	0.31	12.00	7.50
CD @ 5 %	5.58	0.93	36.38	22.74
CV %	5.11	9.26	13.00	13.00

Effect of different methods and levels of application of taegro on diseases incidence of *Alternaria solani* and *Phytophthora infestans* at 65 and 90 DAT

Significant difference in % diseases index of *A. solani* and *Phytophthora infestans* were observed among the treatments at both 65 and 90 DAT (Plate.1). The treatment T_7, *i.e.*, standard chemical control by mancozeb @ 2g/l recorded the lowest % diseases index of *Alternaria solani* of 8.97 and 15.79 PDI at 65 DAT and 90 DAT and *Phytophthora infestans* of 5.13 and 14.69 PDI at 65 DAT and 90 DAT (Table 3). The probable reason for such finding may be that, mancozeb would have affected the spore germination and mycelial development, which may have resulted in the inhibition of disease producing activity of pathogen in the plant and induced resistance in plant. This may be the reason for minimum disease intensity and maximum yield as compared to other treatments. The results are in agreement with the findings of[13], who reported that metalaxyl acts mainly by inhibiting fungal growth and sporulation through the inhibition of RNA synthesis.

TABLE 3. Effect of different methods and levels of taegro as bio-fungicide against *Alternaria solani and Phytophthora infestans* disease incidence at 65 and 90 DAT of hybrid tomato cv. NS -501

Treatments	Percent diseases index of *A. solani*		Percent diseases index of *P. infestans*	
	At 65 DAT	At 90 DAT	At 65 DAT	At 90 DAT
T_1- Seed treatment with taegro @ 4g/kg seed	13.88 ^{bc}	20.49 ^{b}	12.51 ^{c}	21.37 ^{cd}
T_2- Seedling dip with taegro @ 4g/l of water for 25 minutes	13.67 ^{bc}	25.23 ^{c}	12.30 ^{c}	24.21 ^{e}
T_3- Both seed treatment with taegro @ 4g/kg seed and soil drenching twice (*i.e.*, 15 and 30 DAT) @ 250 g/ha	15.02 ^{c}	27.64 ^{c}	14.17 ^{c}	26.59 ^{f}
T_4- Both seedling dip with taegro @ 4g/l of water and soil drenching twice (*i.e.*, 15 and 30 DAT) @ 500 g/ha	13.20 ^{b}	21.82 ^{b}	11.07 ^{b}	23.83 ^{de}
T_5- Both soil drenching and foliar spray with taegro @ 250g/ha on 15 and 30 DAT	12.45 ^{bc}	20.24 ^{b}	9.84 ^{b}	20.44 ^{bc}
T_6- Both soil drenching and foliar spray with taegro @ 500g/ha on 15 and 30 DAT	11.15 ^{ab}	19.46 ^{b}	9.70 ^{b}	18.38 ^{b}
T_7- Standard chemical control by spraying mancozeb @ 2 g/l 10 days interval throughout crop growth period	8.97 ^{a}	15.79 ^{a}	5.13 ^{a}	14.69 ^{a}
T_8- Untreated control	20.03 ^{d}	33.26 ^{d}	18.60 ^{d}	30.60 ^{g}
F test (p=0.05)	*	*	*	*
S Em ±	1.05	0.88	0.69	0.86
CD @ 5 %	3.19	2.67	2.08	2.61
CV %	13.43	6.64	10.18	16.62

DAT: Days after transplanting; In a column means followed by same letter (s) are not significantly different as per DMRT; * Significant at p=0.05 level

Treatments T_1, T_2, T_4, T_5, and T_6 were the next best in recording less % disease index of *A. solani* at both 65 and 90 DAT. The highest % disease index of *A. solani* (20.03 and 33.26) were noticed in T_8 *i.e.,* untreated control. The next best treatments in controlling *Phytophthora* disease were T_4, T_5 and T_6 at 65 DAT and T_5 and T_6 at 90 DAT. Combined application methods of taegro *viz.,* seedling dipping and soil drenching (T_4) or both soil drenching and foliar sprays with either of the concentrations (T_5 and T_6) were relatively more effective in controlling *Phytophthora infestans.*The maximum % disease index (18.60 and 30.60) was observed in treatment T_8, untreated control (Plate.2). The results obtained in this experiment are in accordance with the findings of [14] in tomato by enhancing systemic resistance in tomato seedlings through induction of growth hormones like indole-3-acetic acid (IAA) and gibberellic acid (GA_3) and defense enzymes like peroxidase, polyphenol oxidase and superoxide dismutase. This was in conformity with the findings of [15 -17] in tomato. *B. subtilis* strains could be effective biocontrol agents against soil fungi plant pathogens and could have a potential bio-fertilizer effect, since they stimulated growth and yield of tomato plants[18]. *B. subtilis* B1, B6, B28 and B99 significantly promoted growth and biocontrol activity against *F. oxysporum* f.sp *ciceris* in chickpea. They were observed to produce IAA, HCN and antifungal volatiles among others [19]. It may be due to the influence of plant growth promotion and induced systemic resistance (ISR) in enhancing the disease resistance in tomato plants. This is in confirmation with studies conducted by [20]. The result shows that *Bacillus* spp are very effective biocontrol agents and should be harnessed for further biocontrol applications[21]. Available reports suggest that specific strains of the species *Bacillus amyloliquefaciens, B. subtilis, B. pasteurii, B. cereus, B. pumilus, B. mycoides,* and *B. sphaericus* elicit significant reductions in the incidence or severity of various diseases on diversity of hosts including greenhouse studies or field trials on tomato, bell pepper, muskmelon, watermelon, sugarbeet, tobacco, Arabidopsis species, cucumber, loblolly pine and tropical crops [22].

PLATE 1 : View of tomato plot- Foliar spray with mancozeb @ 2 g/l. (T_7)

PLATE 2: View of tomato plot- Untreated control. (T_8)

The present investigation revealed that, apart from the standard chemical control through mancozeb treatments, the application bio-fungicide (taegro) by both soil drenching and foliar spray @ 500 g per hectare has proved to be the most appropriate and economically viable treatment in enhancing yield and disease control. The next best treatment was soil drenching and foliar spray of taegro @ 250 g per hectare.

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