*Plant Archives* Vol. 17 No. 2, 2017 pp. 935-940 *ISSN 0972-5210*

# MANAGEMENT OF LEAF BLIGHT THROUGH INDUCTION OF DEFENSE ENZYMES IN TOMATO

**M. Magesh[1] and P. Ahila Devi[2]**

[1]Department of Plant Pathology, Annamalai University, Annamalainagar - 608 002 (Tamil Nadu), India.
[2]Plant Pathology Unit, Tamil Nadu Rice Research Institute, Aduthurai - 612 101 (Tamil Nadu), India.

## Abstract

Tomato plants treated with *P. fluorescens* as seed treatment @ (10ml /kg of seed) plus *Allium sativum* as foliar spray @ (15% at 30 & 45 DAT) and challenge inoculated with *A. solani* showed induction of earlier and increased level of defense enzymes *viz.,* PO, PPO, PAL and catalase as well as PR proteins - chitinases and β-1, 3 glucanases. In field trial, the treatment $\mathbf{T}_7$ with application of *P. fluorescens* as seed treatment @ (10ml/kg of seed) plus *Allium sativum* as foliar spray @ (15% at 30 & 45DAT) increased the fruit yield.

***Key words :*** Tomato- late blight, PO-PPO, *P. fluorescens, Alium sativam*.

## Introduction

Tomato (*Lycopersicon esculentum* Mill.) belongs to the family solanaceae is one of the most remunerable and widely grown vegetable in the world. It is cultivated for its fleshy fruit and the area under its cultivation is increasing day by day due to its nutritional value, demand and high yield.

India is the fourth largest producer of tomato globally, contributing around 11.9 MT/ year. However, its average per hectare production is lesser (19.6 MT) when compared to the world average (28.2 MT) (Anonymous, 2014). The yield of tomato is restricted to a great extent due to various diseases and insect pests associated with tomato cultivation. Amongst these, tomato is highly susceptible to early blight, late blight and *Fusarium* wilt (Panthee and Chen, 2010). Fungal infestations cause deterioration in the quality of tomato, minimize yield and fetch less market value.

In India, the yield loss due to early blight disease was estimated to be 10 to 80 per cent (Singh, 1985; Abada *et al*., 2008). Disease management in tomato is widely practiced using chemicals (Singh *et al*., 2001). However, indiscriminate use of chemicals led to the development of fungicidal resistance by the pathogen, environmental pollution and health hazards (Rai *et al.,* 2000). Hence, a search for alternative methods of plant disease management is on increase.

Plants have latent defense mechanism against pathogens, which can be systemically activated upon exposure of plants to stress or infection by pathogens (Baker *et al*., 1997). This phenomenon is called induced systemic resistance (Tuzun and Kuc, 1991). This mechanism operates through the activation of multiple defense compounds at sites distant from the point of pathogen attack (Dean and Kuc, 1985).

Moreover, there is an increased public demand for sustainable and chemical residue-free food production (Arthur, 1996). In response to this, biofungicides, derived either from microbes or plants, emerged as promising alternative strategies. Neem (*Azadirachta indica*), garlic (*Allium sativum*), onion (*Allium cepa)* and few other plants inhibit early blight in potato and tomato (Prasad and Naik, 2003; Mate *et al*., 2005). Leaf extracts of *Prosopis juliflora* and *Cocos nucifera* inhibited proliferation of spores of *Alternaria solani* above 90% (Thiribhuvanamala *et al*., 2001).

Plants provide abundant resource of antimicrobial compounds and have been used for centuries to inhibit microbial growth (Jun-Dong *et al.,* 2006). The extracts of many allelopathic plants are now known to exhibit antimicrobial activities. Flavanoids, triterpenoids, steroids and other phenolic compounds in plants have been reported to have antimicrobial activity (Hostetman *et al*., 1995). Plant Growth Promoting Rhizobacteria (PGPR) combined

with plant extracts for triggering plant 'immune system' to fight against various diseases have been tried (Latha *et al*., 2009).

One of the problems encountered with the use of biocontrol agents is that they may not perform equally well in a variety of environmental conditions. Hence, an integrated approach involving biocontrol agents and plant products would ensure the maximum suppression of early blight disease and higher yield of tomato without any harmful effect on the ecosystem. Therefore, the present study was undertaken to investigate the effect of combined application of PGPR along with an effective plant extract for managing tomato early blight by the induction of defence related enzymes in tomato pre treated with PGPR, plant product and challenge inoculated with *A. solani.*

## Materials and Methods

### Efficacy of *Allium sativum* as foliar spray on the early blight disease incidence of tomato under pot culture

Based on the assessment of the efficacy of the various plant products under *in vitro* condition, *Allium sativum* leaf extract @ 15% conc. which showed maximum reduction on the growth of *A. solani* was further tested at different concentrations against early blight disease in pot culture experiment. The early blight susceptible variety PKM1 grown in earthen pots was used for the study. *Allium sativum* leaf extract @ 15% conc. was sprayed two days after inoculation of the pathogen and second spray was given at fortnightly interval. The crop was maintained in poly house with frequent spraying of water to provide adequate moisture and relative humidity to enable successful infection by the pathogen. The experiment were conducted in a randomized block design with three replications for each treatment and a suitable control. The fungicide Mancozeb 75% WP (0.25%) was used for comparison and the standard agronomic practices recommended by the State Agricultural Department were followed. The observations on PDI was assessed.

### Effect *P.fluorescens* and *Allium sativum* on the management of early blight of tomato under pot culture

#### Foliar spray

Liquid based bioformulations were mixed with the water @ 2 ml $lit^{-1}$ and used as foliar spray at 30$^{th}$ and 45$^{th}$ day after transplanting

#### Chemical spray

Mancozeb @ 0.2% was used for seed treatment and foliar spray were given on 30$^{th}$ and 45$^{th}$ day after transplanting, both in pot culture as well as field experiments. Mancozeb served as chemical check.

#### Glasshouse studies

Glasshouse studies were conducted to test the efficacy of combined application *P.fluorescens* and *Allium sativum* for assessing their influence on the incidence of early blight of tomato.

#### Enzyme extraction

The leaf and root tissues were collected from treated and control tomato plants and immediately extracted with 2 ml of 0.1 M sodium citrate buffer (pH 5.0) at 4°C. The homogenate was centrifuged for 20 min at 10,000 rpm. Protein extracts prepared from tomato tissues were used for estimation of defense enzymes. Sodium phosphate buffer 0.1 M (pH 7.0) was used for the extraction of peroxidase, polyphenol oxidase, catalase and phenylalanine ammonia lyase enzymes.

#### Assay of peroxidase (PO)

Assay of PO (EC 1.11.1.7) activity was carried out as per the procedure described by Hammerschmidt *et al*. (1982). The reaction mixture consisted of 2.5 ml of a mixture containing 0.25 per cent (v/v) guaiacol in 0.01 M sodium phosphate buffer, pH 6.0 and 0.1 M hydrogen peroxide. Enzyme extract (0.1ml) was added to initiate the reaction, which was followed colorimetrically at 470 nm. Crude enzyme preparations were diluted to give changes in absorbance at 470 nm of 0.1 to 0.2 absorbance units/min. The boiled enzyme was used as blank. Activity was expressed as the increase in absorbance at 470 nm $min^{-1}$ $mg^{-1}$ of plant tissue.

#### Assay of polyphenoloxidase (PPO)

One gram of sample was homogenized in 2 ml of 0.1 M sodium phosphate buffer (pH 6.5) at 4$^0$C. The homogenate was centrifuged at 20,000 rpm for 15 min at 4$^0$C. The supernatant served as enzyme source and polyphenoloxidase activity was determined as per the procedure given by Mayer *et al.* (1965). The reaction mixture consisted of 1.5 ml of 0.1 M sodium phosphate buffer (pH 6.5) and 200 µl of the enzyme extract. To start the reaction, 200 µl of 0.01 M catechol was added and the activity was expressed as change in absorbance $min^{-1}mg^{-1}$ of plant tissue.

#### Assay of phenylalanine ammonia-lyase (PAL)

One gram of plant sample was homogenized in 3 ml of ice cold 0.1 M sodium borate buffer, pH 7.0, containing 1.4 mM of 2-mercaptoethanol and 50 mg of insoluble polyvinyl pyrrolidone (PVP). The resulting extract was filtered through cheese cloth and the filtrate was centrifuged at 20,000 rpm for 15 min at 4°C and the

supernatant was used as the enzyme source. PAL activity was determined as the rate of conversion of L-phenylalanine to trans-cinnamic acid at 290 nm. Sample containing 0.4 ml of enzyme extract was incubated with 0.5 ml of 0.1 M borate buffer, pH8.8 and 0.5 ml of 12 mM L -phenylalanine in the same buffer for 30 min at 30°C. The amount of trans-cinnamic acid synthesized was calculated using its extinction coefficient of 9630 $M^{-1}cm^{-1}$ (Dickerson *et al.*, 1984). Enzyme activity was expressed in fresh weight basis as nmol trans-cinnamic acid $min^{-1}$ $mg^{-1}$ of plant tissue.

**Assay of catalase (CAT)**

CAT activity was assayed spectrophotometrically as described by Chaparro-Giraldo *et al*. (2000) using 3 ml assay mixture containing 100 mM potassium phosphate buffer (pH 7.5) and 2.5 mM $H_2O_2$ prepared immediately before use and 100 µl enzyme extract. The activity was measured by monitoring the degradation of $H_2O_2$ using UV-Visible Spectrophotometer (Varian Cary 50) at 240 nm over 1 min, against a plant extract-free blank. The decrease in $H_2O_2$ was followed as the decline in optical density at 240 nm, activity was calculated using the extinction coefficient ($\varepsilon_{240nm}$ = 40 $mM^{-1}$ $cm^{-1}$) for $H_2O_2$ and expressed in mmol $min^{-1}$ $mg^{-1}$ of plant tissue.

**Assay of chitinase**

The colorimetric assay of chitinase was carried out according to the procedure developed by Boller and Mauch (1988). One gram of tomato tissue was extracted with 5 ml of 0.1 M sodium citrate buffer (pH 5.0).

The homogenate was centrifuged for 10 min at 20,000 g at 4°C and the supernatant was used as enzyme source.

## Results and Discussion

Plants are bestowed with various defense related genes. It is well known that the defense genes are sleeping genes and appropriate stimuli or signals are needed to activate them. In the present study the plants treated with *P. fluorescens* @ 10ml /kg as seed treatment and foliar spray with *Allium sativum* @ (15% at 30 & 45 DAT ($T_7$) recorded the maximum activity of defense related enzymes and PR proteins *viz*., peroxidise (PO), polyphenoloxidase (PPO), phenylalanine ammonia lyase (PAL), catalase, chitinase and β-1, 3 glucanases. Meera *et al*., (2013) reported that increase in enzymes PAL, PO, PPO, catalase, chitinase, β-1, 3 glucanases and phenolics in rice plant treated with combination of seed treatment with *P. fluorescens* and foliar spray with *Ecalyptus globules*.

Hence, the enhanced resistance against early blight in tomato plants treated with *P. fluorescens* @ 10ml /kg as seed treatment and foliar spray with *Allium sativum* @ (15% at 30 & 45 DAT ($T_7$)) can be attributed to the direct inhibitory effect as well as their ability to induced systemic resistance against *A. solani.*

**Peroxidase (PO)**

The present study revealed that plants treated with *P. fluorescens* @ 10ml /kg as seed treatment and foliar spray with *Allium sativum* @ (15% at 30 & 45 DAT ($T_7$)) and challenge inoculated with the pathogen showed enhanced stimulation of peroxidiase. The increased PO activity has been correlated with resistance and these enzymes are involved in the polymerization of proteins and lignin or precursor into plant cell wall, thus constructing a physical barrier that could prevent pathogen penetration of cell walls (Bradley *et al*., 1992). Peroxidases have been implicated in the regulation of plant cell elongation, phenol oxidation, polysaccharide cross-linking, IAA oxidation, cross linking of extension monomers, oxidation of hydroxyl–cinnamyl alcohols into free radical intermediates and wound healing (Vidhyasekaran *et al*., 1997a).

**Polyphenol oxidase (PPO)**

The results of the present study indicated that accumulation of polyphenol oxidase was higher in pretreated tomato plants and challenged with early blight pathogen, *A. solani.*

**Phenylalanine ammonia lyase (PAL)**

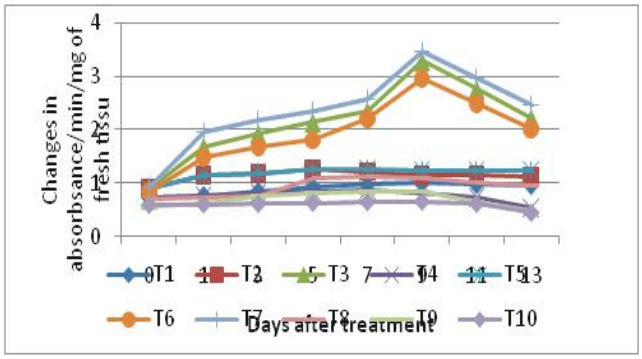
In the present study, reduced disease incidence might be due to increased activity of PAL in plants treated with *P. fluorescens* @ 10ml /kg as seed treatment and foliar spray with *Allium sativum* @ (15% at 30 & 45 DAT ($T_7$)) and challenge inoculated with *A. solani*. The maximum accumulation of PAL upto 9 days constituted for enhancing the resistance in tomato plants against early blight disease.

PAL is the key enzyme in inducing synthesis of salicylic acid (SA), which induces systemic resistance in many plants. In rice, ZB8 PAL gene was found to be induced by the elicitor treatment in rice cells (Li *et al*., 1993).

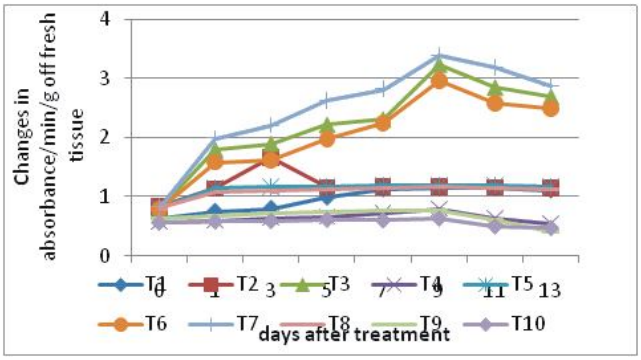
**Catalase**

In the present study, reduced disease incidence might be due to increased activity of catalase in plants treated with *P. fluorescens* @ 10ml /kg as seed treatment and foliar spray with *Allium sativum* @ (15 per cent at 30 & 45 DAT ($T_7$)) and challenge inoculated with *A. solani.* The maximum accumulation catalase activity upto 5 days constituted for enhancing the resistance in tomato plants against early blight disease.

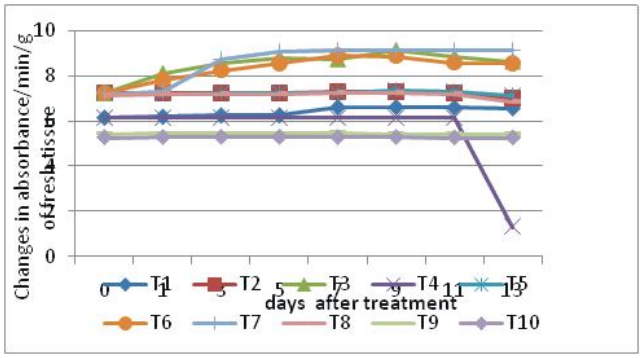
Katsuwon and Anderson (1989) correlated the production of catalase by *P. putida* with the effective rhizosphere colonization. Generally, toxic molecules *viz*., hydrogen peroxide or superoxide is produced from plants during various development stages. To scavenge these



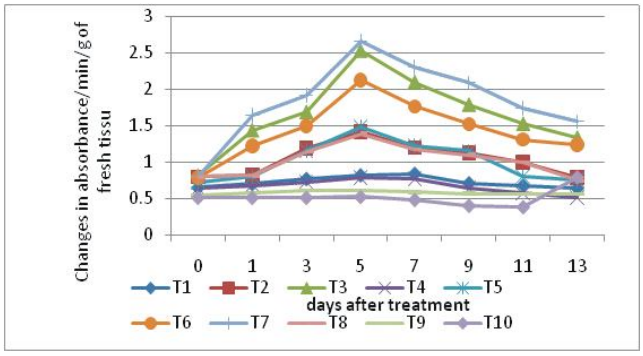

Po



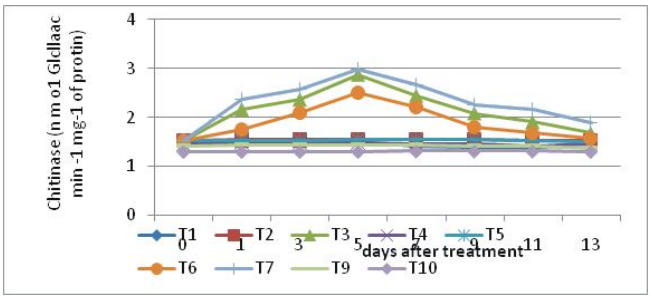

Ppo




Pal




Catalase




Chitinase

toxic molecules, fluorescent pseudomonads were found to produce catalase enzyme to convert hydrogen peroxide and superoxide into diatoic oxygen and water. Also, plants produce active oxygen species (AOS) such as superoxide anion ($O_2$), hydrogen peroxide ($H_2O_2$) and hydroxyl radical (OH) as one of the earliest responses to infection by pathogens Scavengers of active oxygen species like catalase (which catalyzes the decomposition of $H_2O_2$) and suppress the oxidative burst.

**PR proteins – Chitinase and β – 1, 3 Glucanases**

Many PR-proteins induced in plants treated with inducing agents have been shown to be chitinases and β-1, 3 glucanases. The production of chitinases in plants has been suggested to be a part of their defense mechanism against fungal pathogens (Schlumbaum *et al*., 1986). Several earlier workers have reported about the early and enhanced induction of chitinases and β-1, 3 glucanases due to treatment with *P. fluorescens* and challenged with the pathogens (Maurhofer *et al*., 1994).

In the present study, reduced disease incidence might be due to increased activity of chitinase and β-1, 3 glucanase in plants treated with *P. fluorescens* @ 10ml/kg as seed treatment and foliar spray with *Allium sativum* @ (15% at 30 & 45 DAT ($T_7$) and challenge inoculated with *A. solani*. Normally fungal cells contain chitin and glucan as their cell wall constituents. The main mode of action of antagonistic activity of microbes are production of chitinase and β-1, 3 glucanase which act on cell walls of organisms which have chitin or glucan as their cell wall component and also through induced systemic resistance in plant system (Singh *et al.,* 1999).

**Effect *P. Fluorescens* and *Allium sativum* on the management of tomato early blight**

In the field trial application of *P. fluorescens* as seed treatment @ 10ml/kg and foliar spray with *Allium sativum* @ (15% at 45 & 60 DAT ($T_7$) recorded minimum early blight incidence and increase fruit yield.

The result indicated that different plant colonization pattern and different mechanism of disease suppression elicited by the combination of *P. fluorescens* and the extract of *Allium sativum* might have offered greater protection to the tomato crop against the attack of *A. solani* causing early blight disease.

The antifungal activity of *Allium sativum* may be due to the presence of sulphur compounds and allicin present in them (Sehajpal *et al*., 2009). The mechanism

**Table 1 :** Effect of *P. fluorescens* and *Allium sativum* on early blight incidence of tomato (pot culture).

| Treatment | Percent disease index | Percent reduction over control | Plant height(cm) | Fruit yield (g/plant) |
|---|---|---|---|---|
| $T_1$ - Seed treatment with *P. fluorescens* @ 10ml /kg | 35.85 | 34.77 | 70.50 | 196.35 |
| $T_2$ - Foliar spray with *P. fluorescens* @ 0.2% at 30 & 45 DAT | 32.72 | 40.46 | 75.73 | 197.31 |
| $T_3$ - $T_1 + T_2$ | 23.78 | 56.73 | 83.60 | 259.14 |
| $T_4$ - Foliar spray with *Allium sativum* @ 15% at 30 DAT | 37.14 | 32.42 | 69.50 | 195.21 |
| $T_5$ - Foliar spray with *Allium sativum* @ 15% at 30 & 45DAT | 26.86 | 51.12 | 76.42 | 199.46 |
| $T_6$ - $T_1 + T_4$ | 24.62 | 55.20 | 79.62 | 245.11 |
| $T_7$ - $T_1 + T_5$ | 20.14 | 62.68 | 85.55 | 313.52 |
| $T_8$ - Mancozeb (0.25%) as foliar spray at 30 & 45 DAT | 19.60 | 64.33 | 74.85 | 210.78 |
| $T_9$ - Inoculated control | 54.96 | - | 59.20 | 165.76 |

Values in the column followed by same letters not differ significantly by DMRT (p=0.05)

**Table 2 :** Effect *P. fluorescens* and *Allium sativum* on early blight incidence of tomato under field condition (2014).

| Treatments | Percent disease index | Percent reduction over control | Fruit yield (t/ha) |
|---|---|---|---|
| $T_1$ - Seed treatment with *P. fluorescens* @ 10ml /kg | 17.02 | 40.77 | 52.42 |
| $T_2$ - Foliar spray with *P. fluorescens* @ 0.2% at 30 & 45 DAT | 16.86 | 41.33 | 52.65 |
| $T_3$ - $T_1 + T_2$ | 12.61 | 56.12 | 55.09 |
| $T_4$ - Foliar spray with *Allium sativum* @ 15% at 30DAT | 17.14 | 40.36 | 51.92 |
| $T_5$ - Foliar spray with *Allium sativum* @ 15% at 30 & 45 DAT | 15.00 | 47.80 | 53.89 |
| $T_6$ - $T_1 + T_4$ | 13.52 | 52.95 | 54.00 |
| $T_7$ - $T_1 + T_5$ | 10.17 | 62.87 | 59.31 |
| $T_8$ - Mancozeb (0.25%) as foliar spray at 30& 45 DAT | 10.00 | 64.61 | 55.60 |
| $T_9$ - Inoculated control | 28.74 | | 47.00 |

Values in the column followed by same letters not differ significantly by DMRT (p=0.05).

of the action of sulfur compounds towards microorganisms is complex and has not yet been fully explained. It is generally recognised that the antimicrobial action of sulfur compounds depends on their hydrophilic or lipophilic character.

Akila *et al*. (2011) positively correlated the treatment with combination of botanical and bacterial antagonist such as *P. fluorescens* and *B. subtilis* on the induction of defense enzymes against *Fusarium* wilt of banana.

Gangopadhyay *et al.* (2010) reported that the extract of the *C. procera* effectively reduced the incidence of *Alternaria* blight of cumin and increased seed yield. Netam *et al*. (2011) reported that garlic bulb extract used as foliar spray was found most effective showing significantly less disease severity (6.0%) against paddy blast disease in rice.

The results of the present study have clearly revealed that combination of *P. fluorescens* along with *Allium sativum* extract would have exerted a synergism and also different mechanisms of disease control, which certainly enhanced greater disease suppression and increased fruit yield of tomato and improved the consistency of biological control under varied climatic conditions.

## References

Abada, K. A., S. H. Mostafa and R. Mervat (2008). Effect of some chemical salts on suppressing the infection by early blight disease of tomato. *Egyptian J. of Applied Science*, **23** : 47–58.

Akila, R., L. Rajendran, S. Harish, K. Saveetha, T. Raguchander and R. Samiyappan (2011). Combined application of botanical formulations and biocontrol agents for the management of *Fusarium oxysporum* f. sp. *cubense*

causing *Fusarium* wilt in banana. *Biological Control*, **57** :175-183.

Anonymous (2014). 2000-2011tomato statistics EN.aspx. www.novagrim.com

Arthur, F. H. (1996). Grain protectants: current status and prospects for the future. *J. of Stored Products Research*, **32** : 293 - 302.

Baker, B., P. Zambryski, B. Staskaawicz and S. P. Dineshkumar (1997). Signaling in plant-microbe interaction. *Science*, **276** :726-733.

Boller, T. and F. Mauch (1988). Colorimetric assay for chitinase. *Meth. Enzymol*., **161** : 430-435.

Bradley, D. J., P. Kjellborn and C. Lamb (1992). Elicitor and wound induced oxidative cross-linking of a plant cell wall proline-rich protein: A novel, rapid defense response. *Cell*., **70** : 21-30.

Chaparro–Giraldo, A., R. M. Barata, S. M. Chabregas, R. A. Azevedo and M. C. Silva–Filho (2000). Soybean leghemoglobin targeted to potato chloroplasts influences growth and development of transgenic plants. *Plant Cell. Rep*., **19** : 961-965.

Dean, R. A. and J. Kuc (1985). Induced systemic protection in plants. *Trends in BioTechnol*., **3** : 125-129.

Dickerson, D. P., S. F. Pascholati, A. E. Hagerman, L. G. Butler and R. L. Nicholson (1984). Phenylalanine ammonia lyase and hydroxycinnamate: CoA ligase in maize mesocotyls inoculated with *Helminthosporium maydis* or *Helminthosporium carbonum*. *Physiological Plan*., **6(2)** :184-192.

Gangopadyay, S., B. R. Bhakar and S. L. Godara (2010). Effect of botanicals on cumin blight caused by *Alternaria burnsii*. *J. mycol. Pl. Pathol*., **40(4)** : 570-574.

Hammerschmidt, R., E. M. Nuckles and J. Kuc (1982). Association of enhanced peroxidase activity with induced systemic resistance of cucumber to *Colletotrichum lagenarium. Physiol. Plant Pathol*., **20** : 73-82.

Hostetman, A., M. Marston, M. Maillard and M. Hamburger (1995). Proceedings of the phytochemicals society of Europe. *Phytochemistry of Plants used in Traditional Medicine*, pp. : 17-43.

Jun-Dong, Z., Xu Zheng, C. Young- Bing, C. Hai-Sheng, Y. Lan, A. Mao-mao, G. Ping-Hui, W. Yan, J. Xin-Ming and J. Yuan-Ying (2006). Antifungal activities and action mechanisms of compounds form *Tribulus terrestris*.L. J. *Enthanopharmacol*., **103** : 76-84.

Katsuwon, J. and A. J. Anderson (1989). Catalase and superoxide dismutase of root colonizing saprophytic fluorescent pseudomonds. *Applied Environmental Microbiology*, **56** : 3576-3582.

Latha, P., T. Anand, N. Ragupathi, V. Prakasam and R. Samiyappan (2009). Antimicrobial activity of plant extracts and induction of systemic resistance in tomato plants by mixtures of PGPR strains and zimmu leaf extract against *Alternaria solani. Biological Control*, **50** : 85-93.

Li, L., R. Qu, A. Kochko, C. Fauquet and R. N. Beachy (1993). An improved rice transformation system using the biolistic method. *Plant Cell Rep*., **12** : 250-255.

Mate, G. D., V. V. Deshmukh, D. J. Jiotode, N. S. Chore and M. Dikkar (2005). Efficacy of plant products and fungicides on tomato earlyblight caused by *Alternaria solani*. *Research on Crops*, **6** : 349 - 351.

Maurhofer, M., C. Hase, P. Meuwly, J. P. Métraux and G. Defago (1994). Induction of systemic resistance of tobacco to tobacco necrosis virus by the root-colonizing *Pseudomonas flourescens* strain CHAO: Influence of the gacA gene and of pyoverdine production. *Phytopathol*., **84** : 139-146.

Mayer, A. M., E. Harel and R. B. Shaul (1965). Assay of catechol oxidase a critical comparison of methods. *Phytochemistry*, **5** : 783-789.

Meera, T., P. Balabaskar and L. Vengadesh Kumar (2013). Potential of *Pseudomonas fluorescens* and *Eucalyptus globules* extract on inducing defense molecules against rice sheath rot disease. *Ann. Pl. Protec. Sci*., **21(2)** : 315-321.

Netam, R. S., A. N. Bahadur, U. Tiwari and R. K. S. Tiwari (2011). Efficacy of Plant Extracts for the Control of (*Pyricularia grisea*) Blast of Rice under Field Condition of Bastar, Chhattisgarh. *Research Journal of Agricultural Sciences*, **2(2)** : 269-271.

Panthee, D. R. and F. Chen (2010). Genomics of fungal resistance in tomato. *Current Genomics*, **11** : 30 - 39.

Prasad, Y. and M. K. Naik (2003). Evaluation of genotype fungicides and plant extracts against early blight of tomato caused by *Alternaria solani*. *Indian Journal of Plant Protection*, **30(2)** : 49-53.

Rai, N. K., L. Tuli, B. K. Sarma and U. P. Singh (2000). Effect of plant extracts on spore germination of some fungi. *Indian J. Plant Pathol*., **18** : 44-47.

Sehajpal, Anil, Saroj Arora and Parminder Kaur (2009). Evaluation of plant extracts against *Rhizoctonia solani* causing sheath blight of rice. *J. of Pl. Protec. Sci*., **1(1)** : 25-30.

Singh, P. P., Y. C. Shin, C. S. Park and Y. R. Chung (1999). Biological control of *Fusarium* wilt of cucumber by chitinolytic bacteria. *Phytopathology*, **89** : 92-99.

Singh, R.S. (1985). *Disease of vegatables crops*. Oxford and IBH Public Co. New Delhi, pp.441.

Thiribhuvanamala, G., G. Ramkrishnan and D. Sabitha (2001). Effect of plant products against *Alternaria solani*, the early blight pathogen of tomato. *South Indian Horticulture*, **49** : 257 - 260.

Tuzun, S. and J. Kuc (1991). Plant Immunization: An alternative to pesticides for control of plant diseases in greenhouse and field. In: *The biological control of plant diseases*. Ed. J. Bay-Peterson, Food and Fertilizer technology centre, Taiwan. pp. 30-40.

Vidhyasekaran, P., R. Rabindran, M. Muthamilan, K. Nayar, K. Rajappan, N. Subramanian and K. Vasumathi (1997a). Development of powder formulation of *Pseudomonas fluorescens* for control of rice blast. *Plant Pathol*., **46** : 291-297.