Biological Forum – An International Journal **16(2): 110-116(2024)**

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Biochemical Analysis of Early Blight Tolerance in Contrasting Genotypes of Tomato (*Solanum lycopersicum* L.)

Amarnath M.[1*], D. Satish[1], S. Sriram[2], K.S. Shivashankara[3], A.T. Sadashiva[2] and K.V. Ravishankar[3]

[1]Department of Biotechnology and Crop Improvement,
University of Horticultural Sciences, Bagalkot (Karnataka), India.
[2]Division of Crop Protection, ICAR-Indian Institute of Horticultural Research, Bengaluru (Karnataka) India.
[3]Division of Basic Sciences, ICAR-Indian Institute of Horticultural Research, Bengaluru (Karnataka) India.

(Corresponding author: Amarnath M.)*
(Received: 08 December 2023; Revised: 27 December 2023; Accepted: 08 January 2024; Published: 15 February 2024)
(Published by Research Trend)

ABSTRACT: Early blight (EB) is one of the most common and highly destructive tomato diseases. *Alternaria solani* is the fungus that causes EB disease in tomato. Warm and humid weather, especially during the growing season, favors the rapid spread of early blight which leads to foliage damage causing the development of dark lesions and reducing the overall photosynthetic capacity of the plant. Premature Leaf Senescence leads to reduced photosynthesis and premature leaf drop can result in fewer and smaller fruits, ultimately leading to a decrease in overall yield. Biochemical studies on tomato early blight involve the investigation of various biochemical aspects related to the interaction between the tomato plant and the pathogenic fungus *A. solani*. The objective of this study was to look at biochemical changes that occur during disease progression in three different tomato genotypes after artificial inoculation with the early blight pathogen *A. solani*. These genotypes differed significantly in the occurrence of disease on leaves at different time intervals. The biochemical analysis of total sugars, reducing sugars, enzymes, total phenolics (TP) and total flavonoids (TF) showed significant differences due to genotype, disease occurrence and interaction of these two factors. Based on the analysis of results obtained, we conclude that, content of these secondary metabolites could be used as a one of the parameters in the evaluation of degree of tolerance to early blight disease in tomato.

Keywords: *Alternaria solani,* early blight, flavonoids, phenolics.

INTRODUCTION

Tomato (*Solanum lycopersicum* L.) is an important horticultural crop and the second most after potato in the world. It is consumed as a salad and also as cooked vegetable, used in puree, pickle and ketchup. The production of tomato is affected by many biotic and abiotic factors. Among the biotic factors, one of the most important fungal diseases affecting tomato production is early blight (EB), caused by *A. solani.*

EB disease leads to severe defoliation, resulting in reduction of fruit number and size. Several control measures include use of resistant or tolerant cultivars, crop rotation, and eradication of weeds and fungicides application (Jones *et al.*, 2006). Most economical control measure is use of resistant cultivars (Chaerani and Voorrips 2006). Identification of additional sources of resistance could facilitate the development of new resistant cultivars or hybrids (Chaerani *et al.*, 2007).

Early blight resistant lines have a higher and faster induction of PR-proteins, such as chitinase and -1, 3-glucanase, peroxidase, polyphenol oxidase, and phenyalalanine ammonia lyase (PAL), during the infection process than susceptible lines. Catalase activity was found to be increasing in a susceptible strain (Rani and Yasur 2009). Induction of defense proteins makes the plant resistant to pathogen invasion and has been linked to tomato pathogen defense. Secondary plant metabolites associated with early blight resistance include higher total phenolic content (tannin, flavonol, and phenol) in early blight resistant varieties' leaves and stems (Bhatia *et al.*, 1972). Total phenolics, total protein content and activities of the antioxidant enzymes (catalase, peroxidase and polyphenol oxidase) were highly up-regulated in resistant groups than in susceptible groups linked with the induction of resistance against *A. solani* (Awan *et al.*, 2018). In the resistance mechanism of plants, there will be involvement of several biochemical substances (Rao, 1968). The estimations of enzymes *viz.* peroxidase, superoxide dismutase and polyphenol-oxidase indicated the role of these oxidative enzymes in host-pathogen interaction with the pathogen.

Plants evoke a series of general defense reactions, including the production of phytoalexins, antimicrobial proteins and biochemicals upon sensing invading microorganisms (Radhajeyalakshmi *et al.*, 2009). Secondary plant metabolites involved in EB tolerance include a higher total phenolic content (tannin, flavonol and phenol) in leaves and stems of EB resistant varieties

(Bhatia *et al.,* 1972). The induction of phenolic compounds synthesis during pathogen attack was demonstrated in tomato leaf tissue (Pearce *et al.*, 1998). The main objective of this investigation was to examine the role of biochemical compounds which are involved in tolerance to *A. solani* in three selected tomato genotypes varying in their responses to EB.

MATERIAL AND METHODS

Plantmaterial. Three tomato genotypes, one Susceptible (Punjab Chhuhara) and two resistant [Acc No. IIHR-2891(8-3-3) and Arka Alok] belonging to *Solanum lycopersicum* were utilized in this experiment. Seeds are collected from Division of Vegetable Sciences, ICAR-Indian Institute of Horticultural Research (IIHR), Bengaluru, INDIA. The contrasting genotypes used in this were screened for EB resistance as described earlier (Amarnath *et al.,* 2019).

Screening for resistance. An experiment was conducted at playhouse of Division of Basic Sciences, ICAR-Indian Institute of Horticultural Research, Bengaluru, during 2019 with CRD (Completely randomized design) with three biological replications. Sowing for seedlings production in polyhouse was done in seedling trays and twenty-five-day-old seedlings were transplanted in the pots and maintained in polyhouse.

Inoculum of *A. solani* isolation and purification. The source of inoculum was isolated from EB-infected tomato leaves from infected plants from field of Division of Vegetable Sciences, ICAR- Indian Institute of Horticultural Research, Bengaluru, India. Purified culture was used to inoculate, using the tissue segment method (Aneja, 2001). The pathogenicity was confirmed by inoculation and re-isolation. Further, the pathogen was maintained on PDA (Potato Dextrose Agar) slant.

Artificial inoculation of *Alternaria solani* culture. 30-day seedlings were artificially inoculated by spraying *A. solani* culture on 30-day-old seedlings. Three plants from each of the genotypes were used for the experiment. The spore count was adjusted to 4.5 $\times 10^4$ sporangia/ml. Seven days old culture of the pathogen was used to prepare a conidial suspension. The homogenized suspension was used for inoculation on the foliage of each plant. The plants were then covered with polythene bags for 48h to provide suitable moisture and humidity for the growth and development of the pathogen. For control, plants were mock inoculated with spraying distilled water. To estimate biochemical parameters from tomato, leaf tissues were sampled at 0, 1, 6 and 10 days post-inoculation (DPI) of the pathogen.

Extraction and determination of flavonoids

Total phenols. The total phenolics of the leaf extracts were determined using the Folin and Ciocalteu reagent, following the method described by Weidner *et al.* (2000). Sample and standard readings were made using a spectrophotometer (T80+ UV/Vis Spectrophotometer) at 700 nm against the reagent blank. Total phenol activity is expressed in mg/100g fresh weight.

Total flavanoids. Total flavonoid in the 80% methanol extract was determined as per Chun *et al.* (2003) Flavonoids develop a brick red colour with $AlCl_3$ and $NaNO_2$ at alkaline pH. The complex's absorbance is measured at 510 nm. Total flavonoid activity is expressed in mg/100g fresh weight.

Total sugars and reducing sugars

The Nelson-Somogyi method is a well-known and widely used method for determining sugar concentrations. This method (Somogyi, 1952) relies on the reduction of the cupric ion to cuprous, which is then titrated with Nelson's arsenomolybdate reagent to produce a blue colour to be read at 600 nm. Total sugar content is expressed in g/100g fresh weight.

PPO (Polyphenol oxidase) activity. Method described by Selvaraj and Kumar (1995) was used to determine the activity of PPO. Reaction mixture containing of buffer, pyrogallol and enzyme extract and the components of substrate blank, enzyme blank and sample absorbance was measured at 450nm up to 5 min at 1 min interval. PPO activity is expressed as absorbance/min/g fresh weight.

Peroxidase activity. Peroxidise activity was measured using Subhas Chander (1990) method. In this method the rate of formation of OPD (Acceptor O-Phenyl diamine) dehydrogenation product is a measure of the POD activity. The increase in absorbance was measured at 450nm up to 5 min at 1 min interval. The enzyme activity was expressed at units/gm fresh weight.

Super oxide dismutase (SOD) activity. SOD enzyme assay was measured using Du and Bramlage (1994) method. Super oxides generated by the interaction of riboflavin and methionine in presence of light oxidizes Nitro blue tetrazolium chloride (NBT) compound into violet colour. Superoxide dismutase enzyme reduces the oxidation of NBT by removing the superoxides formed during the reaction, thereby reduces the colour development. Therefore, the extent of reduction in the colour development in presence the enzyme is proportional to the enzyme activity. Absorption was measured at 560 nm. Enzyme activity was expressed as units/gm fresh weight.

RESULTS AND DISCUSSION

Complex interaction between host, pathogen, disease stage and crop leads to resistance and susceptibility to a disease. Several biochemical substances are considered to be associated with the mechanism of resistance in plants. Plants can recognize the pathogen infection and activate defense responses leading to host-pathogen interaction by producing first line of defense compounds. Among the defense compounds phenols, flavonoids and sugars that are produced due to the enhanced activities of peroxidase, polyphenol oxidase and super oxide dismutase play major in accumulation and quenching of free radicals in plants during stress period. In the present study the differences in production of biochemical compounds was analyzed between resistant and susceptible tomato genotypes.

Total phenols and flavonoids. Since phenolics are fungi toxic in nature, they may contribute to increasing the mechanical strength of host cell walls while also inhibiting fungal growth. Significant differences between resistant and susceptible genotypes were observed in this study from 0 DPI to 10 DPI. During

disease progression, all three tomato genotypes showed an increasing trend in phenol and flavonoid content. Highest increase of phenols content was recorded in resistant parent IIHR-2891from 55.85 to 128.92 mg/100g FW at 0 DPI to 10 DPI, followed by Arka Alok (58.83 to 123.29 mg/100g FW). Lowest increase in phenol content was recorded in susceptible genotype Punjab Chhuhara from 54.23 to 85.94 mg/100g FW (Fig. 1). (Yao *et al.*, 1995) Changes in the level of phenolic compounds in plants have been shown to change disease susceptibility. Plants with higher phenolic content have been linked to higher pathogen resistance (Velazhahan and Vidhyasekaran 1994). In the present study, the accumulation of phenolics was higher in the resistant genotype than in the susceptible genotype. The production and localization of phenolics is critical in plant-microbe interactions, and this has a significant impact on disease resistance (Taheri and Tarighi 2011). Non-host resistance has been linked to the constitutive expression of phenols, which are thought to function as preformed inhibitors (Nicholson and Hammerschmidt 1992).

Increase in flavonoid content was recorded in resistant line IIHR-2891(8-3-3) ranging from 49.09 to 110.83 mg/100g FW at 0DPI to 10 DPI. Similar trend was observed in Arka Alok (43.99 to 100.15 mg/100g). Susceptible parent Punjab Chhuhara recorded least increase in flavonoid content ranging 0.98 to 63.50 mg/100g FW at 0DPI to 10 DPI (Fig. 2). There was no significant change found in the levels of phenols and flavonoids in mock inoculated samples. In the present study, the accumulation of phenolics and flavonoids was higher in the resistant genotypes than the susceptible genotype (Fig. 2). Tomato leaves produce phenolic compounds as part of their defence system. A high phenolic content level through infected plant parts can have a significant impact on plant health by inhibiting pathogen attachment, invasion, and infection (Agrios, 2005). Phenolic compounds play an important role in plant pathogen defence (Bhattacharya *et al*. 2010; Kurbat *et al.* 2016). The accumulation of phenolics and antioxidant activity in infected leaves is a post-infection response and the first stage of defence. Phenolic compounds have also been identified as the most influential secondary metabolites in the resistance of pearl millet plants to *Sclerospora graminicola*, cowpea to Black eyed cowpea mosaic virus, and raspberry to *Didymella applanata* and *Paraconiothyrium fuckelii* (Arun *et al.*, 2010; Petkovsek *et al*., 2011; Shilpashree *et al.*, 2013). Because phenolics are fungi-toxic in nature, they may contribute to increasing the mechanical strength of host cell walls while also inhibiting fungal growth. It has been demonstrated that changing the level of phenolic compounds in plants changes the degree of disease susceptibility (Yao *et al.* 1995). There is a relation between flavonoid levels in the leaves and resistance to fungal pathogens, particularly *Venturia inaequalis* (Mikulic *et al*., 2011).

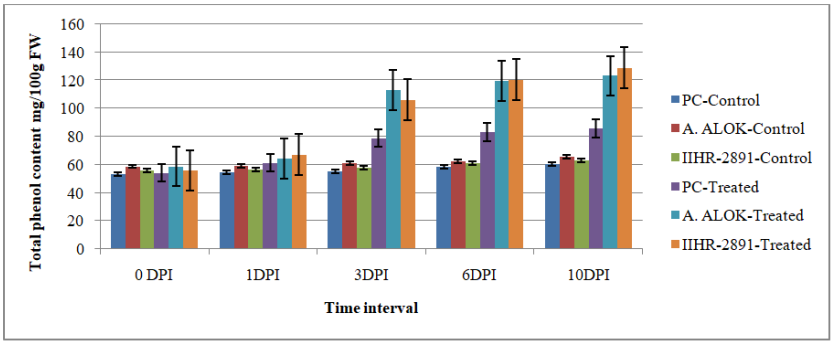

Fig. 1. Total phenol content (mg/100g FW) in susceptible (Punjab Chhuhara) and tolerant (IIHR-2891 and Arka Alok) tomato genotypes following infection with *A. solani* (PC-Punjab Chhuhara, A. Alok- Arka Alok).

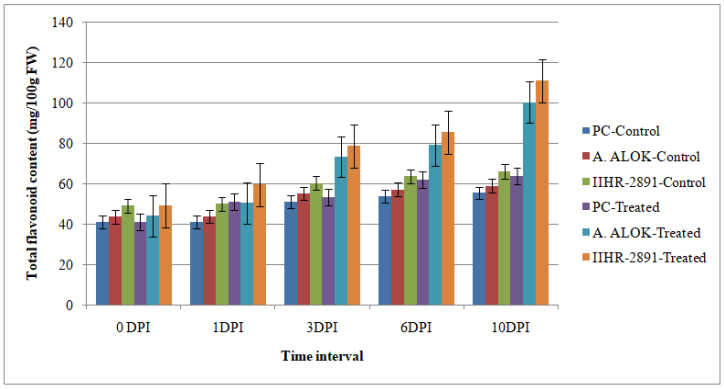

Fig. 2. Total flavonoid content (mg/100g FW) in susceptible (Punjab Chhuhara) and tolerant (IIHR-2891 & Arka Alok) tomato genotypes following infection with *A. solani* (PC-Punjab Chhuhara, A. Alok- Arka Alok).

Total sugars estimation (g/100g FW). Total sugars content was relatively higher in resistant genotypes at earlier stage. After pathogen inoculation recorded gradual decrease in sugar content all the genotypes. Sugar content decreased from 1.7 to 1.2 g/100g in resistant genotype IIHR-2891 followed by Arka Alok

from 1.5 to 1.1 g/100g (Fig. 3). Reducing sugars also shown similar pattern such as total sugars at 0DPI reducing sugar content was high, after pathogen inoculation there was decrease in reducing sugar content (Fig. 4). Variation in the total sugars and reducing sugars was very low in the control samples.

The magnitude of decrease in resistant lines was lower than susceptible lines. Because of their high sugar content, younger leaves are more resistant to EB than older leaves. As a result of the high sugar content and glycol-alkaloids, a late maturing crop may appear EB resistant without even having resistant genes. The sugar content of the leaves decreases as the plant ages increasing the chances of disease development. A decrease in sugar content has been proposed as a cause of increased EB susceptibility in older or weakened leaves and plants (Rotem, 1994).

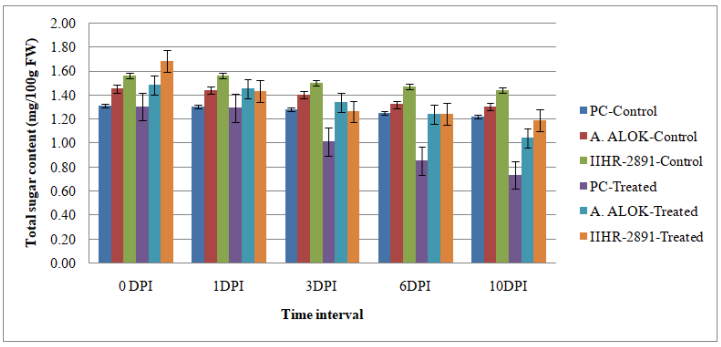

Fig. 3. Total sugar content (mg/100g FW) in susceptible (Punjab Chhuhara) and tolerant (IIHR-2891 & Arka Alok) tomato genotypes following infection with *A. solani* (PC-Punjab Chhuhara, A. alok- Arka Alok).

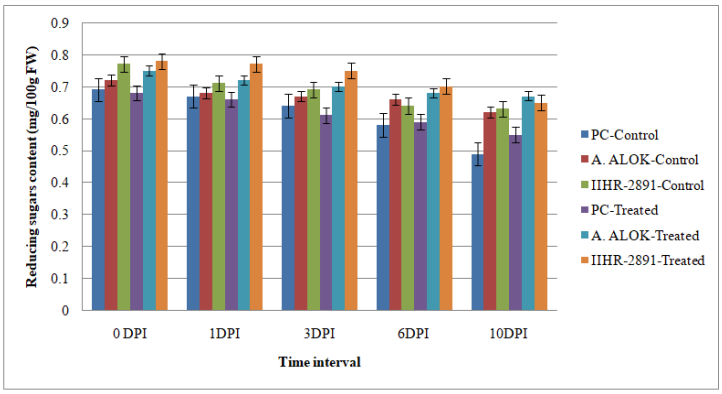

Fig. 4. Reducing sugars content (mg/100g FW) in susceptible (Punjab Chhuhara) and tolerant (IIHR-2891 & Arka Alok) tomato genotypes following infection with *A. solani* (PC-Punjab Chhuhara, A. Alok- Arka Alok).

Peroxidase activity. Enzymatic activity of peroxidase was slightly higher in resistant control compared to susceptible control. Upon infection with *A. solani* pathogen peroxidase activity was enhanced (1.24 U/mg of protein) in resistant genotypes 8-3-3 followed by Arka Alok (0.82) compared to susceptible genotype (0.68 U/mg of protein) at 10DPI. Slight decrease in peroxidase activity was observed in resistant genotype at 3 DPI (Fig. 5).

Peroxidase is important in the production of reactive oxygen species, which are directly toxic to pathogens or indirectly reduce pathogen spread by increasing cross-linking and lignification of plant cell walls (Hammond-Kosack and Jones 1996). The number of resistant interactions involving plant pathogenic fungal and bacterial interactions increased POX activity (Flott *et al.,* 1989; Reimers *et al.,* 1992; Deborah *et al.,* 2001). POX is required for the final polymerization of phenolic derivatives into lignin, as well as for suberization and wound healing (Ward *et al.,* 1991). The results of the present study indicated that peroxidase activity was significantly increased in IIHR-2891and Arka Alok after treatment in EB when compared to Punjab Chhuhara.

Polyphenol oxidase (PPO) activity. The role of polyphenol oxidase in defence against pathogens has been clearly described in host plants. A significant increase in polyphenol oxidase activity was observed in resistant genotype IIHR-2891(0.02) at 10 DPI (Fig. 6). In response to *A. solani* infection, there was systemic up-regulation of Polyphenol oxidase, which was found in upper node leaves but not lower node leaves (Thipyapong and Steffens 1997). PPO is induced by signalling molecules such as jasmonic acid/methyl jasmonate (MeJA), systemin, and salicylic acid in response to mechanical wounding, fungal and bacterial infection (Constabel *et al.,* 2000; Stewart *et al.,* 2001). PPOs catalyse the oxygen-dependent oxidation of o-dihydroxyphenols to o-quinones, which are more pathogen-toxic than the former. Quinones' direct toxicity against pathogens has also been proposed (Mayer *et al.,* 1965). Systemic induction of PPO expression in response to wounding and pathogens may provide an extra line of defence to protect plants from further pathogen and insect attack (Stout *et al.* 1999; Thipyapong *et al.,* 1995).

Superoxide dismutase (SOD). Superoxide dismutase catalyzes the dismutation of superoxide radicals to

oxygen and hydrogen peroxide during stress condition. At 0 days of post inoculation SOD activity was slightly higher in resistant genotypes (IIHR-2891and Arka Alok) compared to susceptible genotype. After infection there was increase in SOD activity in all the lines but the magnitude of increase in resistant lines was higher than susceptible line. The highest SOD activity recorded in IIHR-2891on par with cv. Arka Alok (Fig. 7). The enhanced SOD activity might increase oxidative stress due to increased production of H_2O_2 and it may induce cell dysfunction and death during stress (Tenhaken *et al*., 1995).

The estimation of PPO, POX, and SOD enzymes revealed the importance of these oxidative and hydrolytic enzymes in plant defence against pathogen infection. POX is required for the final polymerization of phenolic derivatives into lignin, as well as for suberization and wound healing (Ward *et al.*, 1991). POX activity was found to be increased in a variety of resistant interactions involving plant pathogenic fungal and bacterial interactions (Deborah *et al.*, 2001; Flott *et al.*, 1989; Reimers *et al.*, 1992). The results of the present study indicated that POX activity significantly increased in Arka Alok and IIHR-2891 after treatment when compared to Punjab Chhuhara.

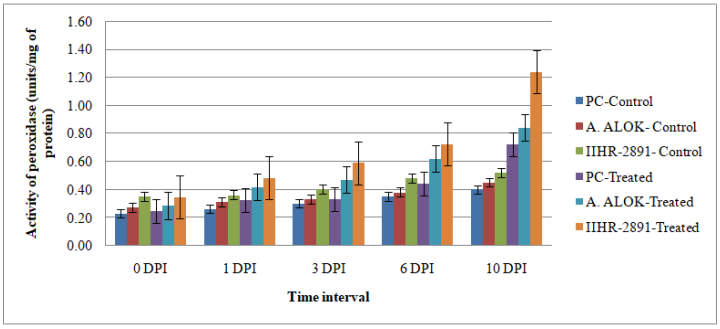

Fig. 5. Activity of peroxidase (units/mg of protein) in tomato genotypes (PC-Punjab Chhuhara, A. Alok- Arka Alok) under control and infected with EB disease.

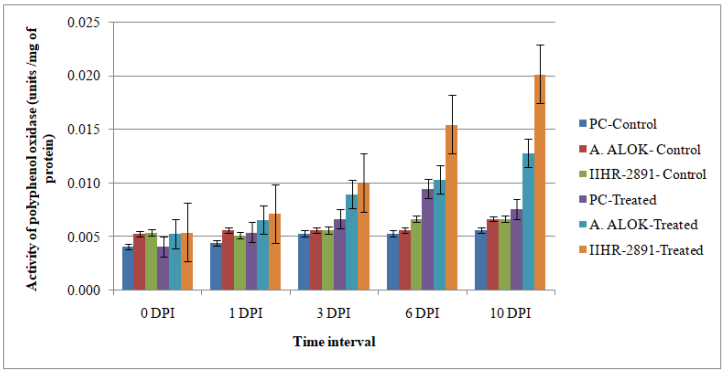

Fig. 6. Activity of polyphenol oxidase (units /mg of protein) in tomato genotypes (PC-Punjab Chhuhara, A. Alok- Arka Alok) under control and infected with EB disease.

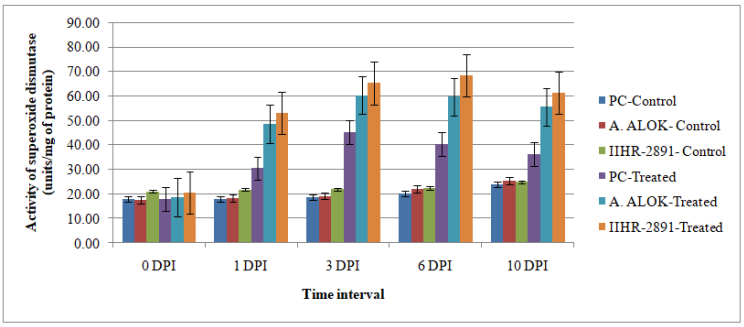

Fig. 7. Activity of superoxide dismutase (units/mg of protein) in tomato genotypes (PC-Punjab Chhuhara, A. Alok- Arka Alok) under control and infected with EB disease.

CONCLUSIONS

The findings revealed that there were significant differences between resistant and susceptible genotypes. PPO, POX, and SOD are three genotypes studied for biochemical compounds such as phenols, flavonoids, total sugars, reducing sugars, and enzymes that play an important role in imparting resistance to tomato EB disease. The content of phenolic acids and flavonoids compounds increased at a faster rate in tolerant genotypes than in susceptible genotypes during disease progression. Furthermore, the genotypes had higher PPO, SOD, and POX activity, indicating antifungal activity as well as higher antioxidant enzyme

activity, which confer tolerance. This phenomenon could be used to evaluate tomato resistance to EB.

Acknowledgement. The authors are very much thankful to UGC-RGNF and ICAR-IIHR, Bengaluru for providing financial support and adequate facilities for the successful completion of this research work.
Conflict of Interest. None.

REFERENCES

Agrios, N. G. (2005). How plants defend themselves against pathogens. In: Plant pathology. 5th ed. *Diego San: Academic Press*, 93–114.

Amarnath, M., Ravishankar, K. V., Satish, D., Sriram, S. and Sadashiva, A. T. (2019). Screening of tomato genotypes against early blight pathogen *Alternaria solani* using detached leaf assay method. Pest Manage *Horticult Ecosyst*, *25*(1), 93–101.

Aneja, K. R. (2001). Experiments in microbiology, plant pathology, tissue culture and mushroom production technology. *New Age International Limited.*

Arun, K., Mali, P. C. and Manga, V. K. (2010). Changes of some phenolic compounds and enzyme activities on infected pearl millet caused by *Sclerospora graminicola*. *Int J Plant Physiol Biochem*, *2*, 6–10.

Bhatia, I. S., Uppal, D. S. and Bajaj, K. L. (1972). Study of phenolic contents of resistant and susceptible varieties of tomato (*Lycopersicum esculentum*) in relation to early blight disease. *Indian Phytopathol*, *25*, 231–235.

Bhattacharya, A., Sood, P. and Citovsky, V. (2010). The roles of plant phenolics in defence and communication during *Agrobacterium* and *Rhizobium* infection. *Mol Plant Pathol.*, *11*, 705–719.

Chaerani, R. and Voorrips, R. E. (2006). Tomato early blight (*Alternaria solani*): the pathogen, genetics, and breeding for resistance. *J Gen Plant Pathol.*, *72*, 335–347

Chaerani, R., Groenwold, R. and Stam, P. (2007). Assessment of early blight (*Alternaria solani*) resistance in tomato using a droplet inoculation method. *J Gen Plant Pathol*., *73*, 96–103.

Chun, O. K., Kim, D. O., Moon, H. Y., Kang, H. G. and Lee, C. Y. (2003). Contribution of individual polyphenolics to total antioxidant capacity of plums. *J Agric Food Chem.*, *51*, 7240-7245.

Constabel, C. P., Yip, L., Patton, J. J. and Christopher, M. E. (2000). Polyphenol oxidase from hybrid poplar. Cloning and expression in response to wounding and herbivory. *Plant Physiol.*, *124*, 285–295.

Deborah, S. D., Palaniswami, A., Vidhyasekaran, P. and Velazhahan, R. (2001). Time course study of the induction of defense enzymes, phenolics and lignin in rice in response to infection by pathogen and non-pathogen. *J Plant Dis Prot*, *108*, 204–216.

Du, Z. and Bramlage, W. J. (1994). Superoxide dismutase activities in senescing apple fruit (*Malus domesticva* Borkh.). *J Food Sci.*, *59*, 581-584.

Flott, B. E., Moersehbacher, B. M. and Reisener, H. (1989). Peroxidase isozyme patterns of resistant and susceptible wheat leaves following stem rust infection. *New Phytol.*, *111*, 413–421.

Hammond-Kosack, K. E. and Jones, J. D. G. (1996). Resistance gene-dependent plant defense responses. *Plant Cell*, *8*, 1773–1791.

Jones, J. B., Jones, J. P., Stall, R. E. and Zitter, T. A.(eds.) (2006). Compendium of Tomato Diseases, *APS Press, St. Paul*, MN.

Mikulic-Petkovsek, M., Slatnar, A., Stampar, F. and Veberic, R. (2009). Accumulation of phenolic compounds in apple in response to infection by the scab pathogen *Venturia inaequalis*. *Physiol Mol Plant Pathol.*, *74*, 60–67.

Mikulic-Petkovsek, M., Slatnar, A., Stampar, F. and Veberic, R. (2011). Phenolic compounds in apple leaves after infection with apple scab. *Biol Plant*, *55*, 725–730.

Nicholson, R. L. and Hammerschmidt, R. (1992). Phenolic compounds and their role in disease resistance. *Annu Rev Phytopathol*, *30*(1), 369-389.

Pearce, G. P., Marchand, P. A., Griswold, J., Lewis, N. G. and Ryan, C. A. (1998). Accumulation of feruloyltyramine and p-coumaroyltyramine in tomato leaves in response to wounding. *Phytochemistry*, *47*(4), 659-664.

Radhajeyalakshmi, R., Velazhahan, R., Samiyappan, R and Doraiswamy, S. (2009). Systemic induction of pathogenesis related proteins (PRs) in *Alternaria solani* elicitor sensitized tomato cells as resistance response. *Sci Res Essays*, *4*(7), 685-689.

Reimers, P. J., Guo, A. and Leach, J. E. (1992). Increased activity of cationic peroxidase associated with an incompatible interaction between *Xanthomonas oryzae* pv oryzae and rice (*Oryza sativa*). *Plant Physiol.*, *99*, 1044–1050.

Rotem, J. (1994). The genus Alternaria biology, epidemiology, and pathogenicity, 1st ed. *The American Phtyopathological Society, St. Paul, Minnesota*, 48-203.

Selvaraj, Y. and Kumar, R. (1989). Studies on fruit softening enzyme and polyphenoloxidase activity in ripening mango (*Mangifera indica* L.) fruit. *J Food Sci Technol.*, *26*, 218–222.

Shilpashree, K., Patil, M. S. and Uma, M. S. (2013). Estimation of total phenols and peroxidase isozyme in plants infected with blackeye cowpea mosaic disease. *Mysore J Agric Sci*., *26*, 318–319.

Somogyi, M. (1952). Notes on sugar determination. *The Journal of Biological Chemistry*, *195*, 19-23.

Stewart, R. J., Sawyer, B. J. B., Bucheli, C. S. and Robinson, S. P. (2001). Polyphenol oxidises induced by chilling and wounding in pineapple. *Aust J Plant Physiol., 28*, 181–191.

Stout, M. J., Fidantsef, A. L., Duffey, S. S. and Bostock, R. M. (1999). Signal interactions in pathogen and insect attack: systemic plant-mediated interactions between pathogens and herbivores of the tomato, *Lycopersicon esculentum*. *Physiol Mol Plant Pathol.*, *54*, 115–130.

Subhas Chander, M. (1990). Enzymatic properties association with resistance to rust and powdery mildew in peas. *Indian J. Hortic.*, *47*, 341-345.

Taheri, P. and Tarighi, S. A. (2011). Survey on basal resistance and riboflavin-induced defense responses of sugar beet against *Rhizoctonia solani*. *J Plant Physiol.*, *168*(10), 1114-1122.

Tenhaken, R., Levine, A., Brisson, L. F., Dixon, R. A. and Lamb, C. (1995). Function of the oxidative burst in hypersensitive disease resistance. *Proc Natl Acad Sci*., *USA92*, 4158– 4163.

Thipyapong, P. and Steffens, J. C. (1997). Tomato polyphenol oxidase: differential response of the polyphenol oxidase F promoter to injuries and wound signals. *Plant Physiol.*, *115*, 409–418.

Thipyapong, P., Hunt, M. D. and Steffens, J. C. (1995). Systemic wound induction of potato (*Solanum tuberosum*) polyphenol oxidase. *Phytochemistry, 40*, 673–676.

Velazhahan, R. and Vidhyasekaran, P. (1994). Role of phenolic compounds, peroxidase and polyphenol oxidase in resistance of groundnut to rust. *Acta Phytopathol Entomol Hung*, *29*, 23–29.

Ward, E., Uknes, S. J., Williams, S. C., Dincher, S. S., Wiederhold, D. L. and Alexander, D. C. (1991). Coordinate gene activity in response to agents that induce systemic acquired resistance. *Plant Cell, 3*, 1085–1094.

Weidner, S., Amarowicz, R., Karamac, M. and Frtczek, E. (2000). Changes in endogenous phenolic acids during development of secale cereal caryopses and after dehydration treatment of unripe rye grains. *Plant Physiol Biochem., 38*, 595-602.

Yao, K., De Luca, V. and Brisson, N. (1995). Creation of a metabolic sink for tryptophan alters the phenyl propanoid pathway and the susceptibility of potato to *Phytophthora infestans*. *Plant Cell, 7*, 1787–1799.

How to cite this article: Amarnath M., D. Satish, S. Sriram, K.S. Shivashankara, A.T. Sadashiva and K.V. Ravishankar (2024). Biochemical Analysis of Early Blight Tolerance in Contrasting Genotypes of Tomato (*Solanum lycopersicum* L.). *Biological Forum – An International Journal, 16*(2): 110-116.