



BACTERIAL WILT OF BRINJAL AND ITS CONTROL

MAISURIYA K¹, UPADHAYAY D², MARCHAWALA F², BHATTACHARYA I² AND
ANDHARE P^{2*}

1: Student, M. Sc. Microbiology, Parul Institute of Applied Sciences, Parul University, Post Limda,
Waghodia, Gujarat, 391760

2: Assistant Professor, Parul Institute of Applied Sciences, Parul University, Post Limda, Waghodia,
Gujarat, 391760

*Corresponding Author: E Mail: Dr. Prasad Andhare: prasad.andhare82145@paruluniversity.ac.in; Tel:
+918200614350

Received 19th Jan. 2021; Revised 20th Feb. 2021; Accepted 19th March 2021; Available online 1st April 2021

<https://doi.org/10.31032/IJBPAS/2021/10.4.1006>

ABSTRACT

Brinjal or eggplant (*Solanum melongena L.*) is an important solanaceous vegetable yield mostly grown in India. It is an associate of the Solanaceae family. Bacterial wilt caused by *Ralstonia solanacearum* is one of the most harmful plant diseases worldwide and deadly effects on many economically important yields. In India, brinjal is the 4th important vegetable grown after potato, onion, and tomato fifteen brinjal accessions were separated in the sickbed pre-inoculated with *R. solanacearum*. Resistant and moderately susceptible accessions exposed a longer incubation period. It is a soil-borne pathogen, Gram-negative, rod-shaped, aerobic, and non-spore-forming bacteria. *R. solanacearum* first charge intercellular spaces of roots where it multiplies before entering xylem vessels and producing exopolysaccharide (EPS), evoking wilt of the infected plant. This review paper focused on recent enhancements in control measures, as well as the bio-control ability and suppression mechanisms. Biological control agents (BCAs) have been controlled by bacteria (90%) and fungi (10%). Avirulent strains of *R. solanacearum*, *Pseudomonas spp.*, *Bacillus spp.* and *Streptomyces spp.* are close to BCAs. Inoculation methods for biological control ability, such as drenching of soil, dipping of roots, and seed coating. Biological control of soil-borne plant pathogens by the addition of vertisol soil either artificially or naturally.

Keywords: Brinjal, Bacterial Wilt, Exopolysaccharide (EPS), *Ralstonia solanacearum*, Biological control agents (BCAs)

INTRODUCTION

Solanum melongena L. is special cultivated solanaceous species that arise from the past. It is recognized as brinjal in India. Brinjal or eggplant (*Solanum melongena L.*) is a significant solanaceous vegetable yield mainly grown in the tropical and subtropical regions of the world. It is of much importance as a warm-weather vegetable yield of the Far East being grown broadly in India, China, Pakistan, and the Philippines. In India, it is one of the most common, suitable, and important vegetable yields grown all over the country [1]. It is a valuable source of minerals and vitamins, in total, its nutritional value is related to other vegetables, and has several medicinal properties [2]. Brinjal arises from Asia is one of the best worldwide vegetables in the world. The name eggplant derives from the shape of the fruit of certain varieties, which are white and shaped very equally to chicken eggs. The color, size, shape of the

brinjal change significantly with the type of cultivar [3]. The plant is thick and grows to a height of 60-120 cm; the leaves are bulky and arranged alternately on the stems. The flowers are huge and violet-colored. Brinjal fruits are known for their presence low in calories and have a mineral composition advantageous for human fitness. They are also an abundant source of potassium, magnesium, calcium. It is implanted in three seasons; first in Kharif (June-September), second in Rabi (November-February), and third in March. This vegetable crop is mostly grown by small and marginal farmers and it is an important source of income for them. Brinjal manufacture faces several problems which cause vast yield losses. Fruit and shoot borer (FSB) is the most devastating insect-pest of brinjal, which causes 60-70% yield loss.

Diseases of brinjal (Table 1)

Table 1: Diseases of brinjal with their causative agent's name

Sr. No.	Disease Name	Causative agent	Reference
1	Damping off	<i>Pythium spp., Phytophthora spp., Rhizoctonia spp., Sclerotium spp. and Sclerotinia spp.</i>	Singh et al., 2014 [4]
2	Phomopsis blight and fruit rot	<i>Phomopsis vexans</i>	
3	Little leaf of brinjal		
4	Bacterial Wilt	<i>Pseudomonas solanacearum</i>	
5	Leaf spot	<i>Cercosporamelongenae</i>	
7	Leaf spot	<i>Alternaria melongenae, Alternaria solani</i>	
8	Fruit rot	<i>Phytophthora nicotianae</i>	
9	Verticillium wilt	<i>Verticillium dahlia</i>	
10	Collar rot	<i>Sclerotium rolfsi</i>	
11	Mosaic	Tobacco mosaic virus	
12	Fusarium		

Disease incidence and Frequency:

The gap between quadrat to quadrat depended upon the size of the measured fields [5]. The Vital quantitative analysis like disease incidence and frequency of broomrapes were calculated using the following methods as recommended by Misra (1968) [6].

$$\begin{aligned} & \% \text{ Wilt incidence} \\ & = \frac{\text{Number of wilted plants in each field}}{\text{Total number of plants in each field}} \times 100 \end{aligned}$$

$$\begin{aligned} & \text{Frequency (\%)} \\ & = \frac{\text{Number of isolation}}{\text{Total number of samples plated}} \times 100 \end{aligned}$$

The pathogen (*Ralstonia solanacearum*):

Ralstonia solanacearum inflicting bacterial wilt in brinjal has been identified as an emerging threat to brinjal cultivation. *R. solanacearum* is a soil-borne, Gram-negative, rod-shaped, aerobic, and non-spore-forming microorganism. It happens wide in tropical, subtropical, and warm temperate regions of the domain and causes significant yield loss in major crops like tomato, eggplant, potato, tobacco, banana, ginger, etc. [7].

Strains of *R. solanacearum* amendment in keeping with host range, geographical distribution, pathogenicity, and physiological properties. Standard on host range, pathogenicity, and colony form on Triphenyl tetrazolium chloride (TTC) medium. Strains of *R. solanacearum* sorted into 5 races (Race 1 to 5) developed on the

aptitude of microorganism to infect a wide host range and build a hypersensitive response on tobacco and 6 biovars (Biovar 1 to 6) developed on the nutritional provides of the microorganism, particularly carbohydrates and organic acids. Strains belonging to Race-1 have a wide host range and pathogenic on different solanaceous family plants and weed hosts. Race-2 is restricted to triploid bananas and Heliconia. Race-3 infects potatoes, whereas Race-4 taints ginger and Race-5 is aggressive specifically on mulberry [8].

Morphological and physiological characteristics of *R. solanacearum*:

The characteristics of *Ralstonia solanacearum* is gram-negative, non-spore-forming, motile rod regarding 0.5-0.7 $\mu\text{m} \times$ 1.5-2.0 μm , an aerobic obligate organism, and is non-encapsulated [9]. It is catalase-positive, oxidase-positive, and nitrate reduction positive, sugars like glucose, maltose, fructose, dextrose, sucrose, mannitol, lactose positive. The *R. solanacearum* unable to hydrolysis starch and gelatin. In broth culture, the organism is inhibited by concentrations of sodium chloride (NaCl) greater than 2%. Liquid and solid (agar) growth media are usually used for culturing the microorganism.

On solid agar medium, separate colonies are usually visible after 36 to 48 hours of growth at 28°C, and triphenyl tetrazolium

chloride (TTC) agar is commonly used for its isolation [10]. After two days on TTC medium, virulent wild-type colonies are large, raised, fluidal, and either entirely white or with a pale red center. For the best strain's growth temperature is 28-32°C. Though, some strains that are pathogenic on potatoes have an under optimal growth temperature of 27°C.

Disease cycle & Host range and geographical distribution:

The causative agent enters the xylem vessels (vascular tissue) of roots and spreads into the stem anywhere it multiplies and wilts by excessive exopolysaccharide production. A lot of its key extra cytoplasmic virulence and pathogenicity factors are transcriptionally measured by a broad network of distinct, interacting signal transduction pathways [11]. Its move in on the plants roots by swimming and attaching to the roots, then clusters on the xylem vessels and block the system by exceptionally secreting cell-wall-degrading enzymes and extracellular polysaccharides (EPS) inside the plant part, ultimately inducing host death. It transmits to near plants through root contact, water source, or human or machinery contact. After cut ends of the wilted plant stem or root are placed in water, cloudy white ooze can be seen [12].

Host range and geographical distribution:

R. solanacearum exists as a broadly distributed causative agent in tropical, subtropical, and some high warmth regions of the world. The microorganism wilt disease has been described and causal agent isolated, in addition to a family of 200 plant species belonging to 50 different botanical families. The disease has a worldwide spreading. This bizarrely wide host range is unremittingly increasing, that the description of the new host is common. The most significant dominant hosts are banana, tomato, and groundnut. The bulk of them ordinarily exists in the solanaceous families [13].

Economic importance of the bacterial wilt disease:

Soil-borne diseases are well-thought-out to be more limiting than seed-borne or air-borne diseases within the production of many crops and account for 10–20% of yield losses annually. *R. solanacearum* is classified as the next most important bacterial pathogen that causes severe yield losses on various solanaceous crops in different parts of the world. 70% brinjal in India and changeable degree losses in many brinjal growing countries of the domain [14].

Epidemiology and survival of the pathogen:

Ralstonia solanacearum enters roots through lesions made by transplanting, cultivation, insects, or certain nematodes and through natural lesions where secondary roots emerge. After classified the host, the bacterium has an attraction for the vascular system, where it increases quickly, substantial the xylem through bacterial cells and slime [15]. After infection, it similarly transfers through the vascular system, the xylem, and lastly blocks water passage, which causes drooping. Typical signs of bacterial wilt can saw limited days after infection, such as wilting and later yellowing, dwarfing, and lastly irreversible, quick wilting and death of plants [16].

Pathogenicity Test:

Pathogenicity test of the pathogen was shown in vitro by sick soil method in plastic pot culture, seeding susceptible local brinjal cultivar under screen house condition. Around two weeks later when

the plants were 10-15 cm long and had adequate roots, they were transplanted in 3.5-inch diameter pots. 50 ml solution of fertilizer (N-P-K: 6-7-22) was applied at the time of planting [17]. Plants watered daily, except for 1 day before inoculation. For inoculation, 2 weeks after transfer, the plant rootstock was spoiled with a scalpel, and 40 ml of the microbial suspension was poured into the soil. The wilt frequency was recorded twice a week after the first wilting symptoms began to appear. The result on the pathogenicity test shown that the test pathogen *R.solanacearum* caused considerably reduced seed germination (55%), maximum pre-emergence (45%), and post-emergence seedling mortality (40%) as compared to uninoculated control germination. 100% pre-emergence seed rot over post-emergence mortality (12%). Firstly, infected seedlings exhibited discoloration, softening of collar region, tumbling, and finally, diseased seedlings died [18] (Table 2).

Table 2: Severity scale of bacterial wilt

Scale	% of shoot wilted	Reference
0	No signs	Mwangi et al., 2011 [19]
1	One leaf wilting (1% - 25%)	
2	2 or 3 leaves wilting (26% - 49%)	
3	Half plant wilting (50% - 74%)	
4	Totally leaves wilting (75% - 100%)	
5	Plant dead	

Evaluation of biocontrol against the pathogen:

Trichoderma a genus of asexually reproducing fungi that are broadly

distributed in nearly all temperate and tropical soils. They are strong adaptable attackers, fast-growing, prolific producers of spores and powerful antibiotic

producers. These properties make these fungi ecologically very effective [20]. The appearance of a distinctive level of genetic diversity and can be familiar to produce a wider range of commercial products of industrial and horticultural importance much of the known biology and many of the benefits of these fungi have been familiar recently.

Trichoderma species produce altered volatile and non-volatile combinations which inhibit the development of phytopathogens. The implement of antibiosis starts a much more complex system, leading to the occurrence of biological control. Between these antibiotics, the assembly of gliovirin, gliotoxin, viridin, pyrones, peptabiols and others have been defined broadly. The production of a huge variety of volatile secondary metabolites by *Trichoderma* (e.g. ethylene, hydrogen cyanide, alcohols, aldehydes, and ketones up to C4 chain-length) also shows an important role in biocontrol. Another antibiotic compound, i.e. peptabiols exhibit antibacterial and antifungal properties, signified byalamethicin [21].

Take discs of 5 mm diameter were occupied after the actively growing colonies of the pathogen *R. solanacearum* and antagonists with the help of a sterilized cork borer. The disc of the pathogen was placed on one side of poured on nutrient

agar plates aseptically though, the discs of antagonists were placed on the opposite side of the pathogen in the same Petri plates and the control was also maintained. The testing was accomplished in three replications. The Petri plates were incubated at 25 + 10°C. Next 7 days of incubation, the apparatus of the interface was detected [22] and the data were verified as percent inhibition through the following formula:

$$\text{Per cent growth inhibition over control} = \frac{dc-dt}{dc} \times 100$$

Where, dc = colony diameter in control

dt = colony diameter in treatment

Bio-control Agents and Their Mechanism of Action in the Controlling of Plant Pathogens:

Bio-control agents involve a baffling range of mechanisms in achieving disease control. The mechanisms employed through the bio-control agents in controlling plant infections are mostly classified into direct and indirect antagonisms.

Direct Antagonism:

Direct antagonism outcomes from the physical interaction and/or high grade of selectivity for the pathogens through bio-control agent. This includes:

Production of antibiotics:

Many PGPR (Plant Growth Promoting Rhizobacteria) can produce peptide antibiotics. These are oligopeptides that prevent the combination of pathogens cell

walls, influence membrane structures of cells and inhibit the formation of initiation complex on the minor subunit of ribosomes. Diversity of antibiotics has been identified, containing compounds produced by *Bacillus*, *Streptomyces*, and *Stenotrophomonas spp.* [23].

Greater than 12 antibiotics are produced by *B. subtilis* strains. The antibiotics formed by most *Bacillus spp.* are active against both Gram-positive and Gram-negative bacteria and pathogenic fungi. Several studies presented an active effect of bacterial antibiotics in the modulation of the resistance system of the plant [24].

Indirect antagonism:

Phosphate solubilization:

Phosphorus is the second greatest important plant growth-limiting nutrient after nitrogen. Most phosphorus is in its insoluble form, although the plants can only absorb phosphorus when it's attached with oxygen as in monobasic ($H_2PO_4^-$) and dibasic forms (HPO_4^{2-}) [25]. To overcome phosphate insufficiency, phosphate dense fertilizers are useful to yields regularly. However, plants can only absorb a limited number of phosphates and the rest is rapidly converted into insoluble P. There is also widespread loss of phosphates in agricultural lands via runoff and much of the phosphate ends up in water reservoirs. Some microorganisms relate to hydrolyzing organic through phosphatase enzymes and

solubilize and inorganic phosphates through organic acid invention. The latter is known as phosphate-solubilizing bacteria. Since more than 90% of bacteria, they are known as Phosphate solubilizing bacteria (PSB) [26].

CONCLUSION

Brinjal is a widespread yield. Bacterial wilt affected by *Ralstonia solanacearum*, is some damages to various important yields, mostly Solanaceous plants, this microorganism is a pathogen of bacterial wilt. Clear thoughtful of pathological & physiological wilt and in pathological wilt – Fungal and Bacterial wilt benefits farming community to follow appropriate & proper control measures. Bacterial wilt recent a major problem in solanaceous crops similar to the potato, tomato, brinjal, pepper. Towards *Ralstonia solanacearum* exists complex nature, varied host range and earlier adaptableness to changing environments stabilizing this shattering plant pathogen have developed extensive challenges. There is an absence effective mode of managing pathogen however, the integrative application controls give the best probable effects. Conceiving technically improved, socially acceptable, farmer-friendly, economically possible, health & environment benign elucidations is an abundant challenge to the global scientific community.

ACKNOWLEDGEMENT

It is our privilege and honour to express our sincerest gratitude to the Parul University, Vadodara, Gujarat for providing me all the necessary support and facilities including state of the art infrastructural facilities with advanced technological scientific laboratories and everything else that was required to carry out this work.

REFERENCES

- [1] Ansar, M., & Ghatak, A. (Eds.). (2019). *the Vegetable Pathosystem: Ecology, Disease Mechanism, and Management*. CRC Press.
- [2] Kumar, G., Meena, B. L., Kar, R., Tiwari, S. K., Gangopadhyay, K. K., Bisht, I. S., & Mahajan, R. (2008). Morphological diversity in brinjal (*Solanum melongena* L.) germplasm accessions. *Plant Genetic Resources*, 6(3), 232.
- [3] Kandoliya, U. K., Bajaniya, V. K., Bhadja, N. K., Bodar, N. P., & Golakiya, B. A. (2015). Antioxidant and nutritional components of eggplant (*Solanum melongena* L.) fruit grown in Saurashtra region. *International Journal of Current Microbiology and Applied Sciences*, 4(2), 806-813.
- [4] Singh, B. K., Singh, S. S. B. K., & Yadav, S. M. (2014). Some important plant pathogenic disease of brinjal (*Solanum melongena* L.) and their management. *Plant Pathology Journal (Faisalabad)*, 13(3), 208-213.
- [5] Zhang, H., Sun, Y., Chang, L., Qin, Y., Chen, J., Qin, Y. & Wang, Y. (2018). Estimation of grassland canopy height and aboveground biomass at the quadrat scale using unmanned aerial vehicle. *Remote sensing*, 10(6), 851.
- [6] Akhter, G., & Khan, T. A. (2020). Survey of parasitic weeds (*Orobanche* spp.) associated with brinjal (*Solanum melongena*) in banda district of uttar pradesh India. *Pakistan Journal of Weed Science Research*, 26(1).
- [7] Thakur, H., Sharma, A., Sharma, P., & Rana, R. S. (2020). An insight into the problem of bacterial wilt in *Capsicum* spp. with special reference to India. *Crop Protection*, 105420.
- [8] Anitha, M., Paranidharan, V., & Karthikeyan, M. (2018). Characterization of *Ralstonia solanacearum* (Smith) Race1, Causing Bacterial Wilt of Brinjal. *Madras Agricultural Journal*, 105(September (7-9)), 306-312.
- [9] Sultana, N. (2016). Characterization of indigenous bio-control agents against wilt complex pathogens of

- tomato (Doctoral dissertation, University of Dhaka).
- [10] Tindall, B. J., Sikorski, J., Smibert, R. A., & Krieg, N. R. (2007). Phenotypic characterization and the principles of comparative systematics. *Methods for general and molecular microbiology*, 330-393.
- [11] Schell, M. A. (2000). Control of virulence and pathogenicity genes of *Ralstonia solanacearum* by an elaborate sensory network. *Annual review of phytopathology*, 38(1), 263-292.
- [12] Sarfo, N. Y. (2018). Importance, Source and Control of Bacteria Wilt Disease in Greenhouse Tomato (*Solanum lycopersicum* L.) in Southern Ghana (Doctoral dissertation, University of Ghana).
- [13] Álvarez, B., Biosca, E. G., & López, M. M. (2010). On the life of *Ralstonia solanacearum*, a destructive bacterial plant pathogen. *Current research, technology and education topics in applied microbiology and microbial biotechnology*, 1, 267-279.
- [14] Ashok, K. R., Chinnadurai, M., Raj, S. V., & Sanjeevikumar, A. (2019). Socio-economic Assessment of LMOs: An Ex ante Analysis of Insect Resistance and Herbicide Tolerance in Maize and Brinjal in Tamil Nadu. In *Socio-Economic Impact Assessment of Genetically Modified Crops* (pp. 101-120). Springer, Singapore.
- [15] Van der Wolf, J. M., & De Boer, S. H. (2007). Bacterial pathogens of potato. In *Potato biology and biotechnology* (pp. 595-617). Elsevier Science BV.
- [16] Caudwell, A., Martelli, G. P., & Agent, C. (1993). Alternate Hosts Natural Spread. *Graft-transmissible Diseases of Grapevines: Handbook for Detection and Diagnosis*, 97.
- [17] Gutarra, L., Herrera, J., Fernandez, E., Kreuze, J., & Lindqvist-Kreuze, H. (2017). Diversity, pathogenicity, and current occurrence of bacterial wilt bacterium *Ralstonia solanacearum* in Peru. *Frontiers in plant science*, 8, 1221.
- [18] Liu, Q. (2015). Bacterial wilt of cucurbits: Impact of plant age on symptom progression and pathogen movement and locating genes associated with host preference and pathogenesis in *E. tracheiphila*.
- [19] Mwangi, M. W., Monda, E. O., Okoth, S. A., & Jefwa, J. M.

- (2011). Inoculation of tomato seedlings with *Trichoderma harzianum* and arbuscular mycorrhizal fungi and their effect on growth and control of wilt in tomato seedlings. *Brazilian Journal of Microbiology*, 42(2), 508-513.
- [20] Alelign, S. (2020). Evaluation of the efficacy of *Trichoderma* and *Pseudomonas* species against bacterial wilt (*Ralstonia* isolates) of tomato (*Lycopersicum* spp.) (Doctoral dissertation, Addis Ababa University).
- [21] Silva, R. N., Monteiro, V. N., Steindorff, A. S., Gomes, E. V., Noronha, E. F., & Ulhoa, C. J. (2019). *Fungal Biology*. Singh, H. B., & Singh, D. P. (2009). From biological control to bioactive metabolites: prospects with *Trichoderma* for safe human food. *Pertanika J Trop Agric Sci*, 32(1), 99-110.
- [22] Kariuki, C. (2020). Evaluation of bacillus and trichoderma species for Biological control of bacterial wilt caused by *ralstonia solanacearum* in tomato (Doctoral dissertation, University of Nairobi).
- [23] Ankati, S., & Podie, A. R. (2018). Understanding plant-beneficial microbe interactions for sustainable agriculture. *Journal of Spices and Aromatic Crops*, 27(2), 93-105.
- [24] Choudhary, D. K., & Johri, B. N. (2009). Interactions of *Bacillus* spp. and plants—with special reference to induced systemic resistance (ISR). *Microbiological research*, 164(5), 493-513.
- [25] Muthukumar, A., Udhayakumar, R., & Naveenkumar, R. (2016). Eco friendly management of damping-off of solanaceous crops caused by *Pythium* species. In *Current Trends in Plant Disease Diagnostics and Management Practices* (pp. 49-90). Springer, Cham.
- [26] Kunwar, V. S., Lamichhane, J., & Gauchan, D. P. (2018). Strategies to improve phosphorus availability in a sustainable agricultural system. *Int. J. Innov. Sci. Res. Technol*, 3(9), 323-331.