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Assessment of seed quality parameters in fruit rot infected seeds of brinjal caused by *Phomopsis vexans* and development of eco-friendly management for quality seed production

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ABSTRACT

Phomopsis vexans is an important seed-borne pathogen and it is highly detrimental to brinjal crop. Among seventeen brinjal genotypes studied for the effect of fruit rot on the seed quality parameters using infected and healthy seeds, the germination percentage of infected seeds is relatively lower than the healthy seeds. The reduction in germination percentage of diseased seeds varied between 14 to 67%. Seed quality parameters like germination and vigour were negatively correlated and the electrical conductivity was positively correlated with fruit rot incidence. This shows that a higher incidence of pathogens will lower the viability and vigour of seeds and the infection level is also dependent on genotype and the environmental condition. The three known biocontrol agents were analyzed by using the dual culture technique for the antagonistic effect on the pathogens. The results showed that out of three biocontrol agents, *Trichoderma harzianum* showed maximum antagonistic effect followed by *Pseudomonas fluorescence* and *Bacillus subtilis.* Different modes of action like mycoparasitism, competition, antibiosis and production of antibiotics and secondary metabolites like harzianiol may act as inhibitors against *Phomopsis vexans*.

Key words: *Brinjal,* Phomopsis vexans*, Seed quality, Biocontrol agents, Antagonistic effect.*

Introduction

Brinjal (*Solanum melongena* L.) is one of the important vegetable crops own all over the world. The name brinjal is popular in Indian subcontinent and is derived from Arabic and Sanskrit, whereas the name eggplant has been derived from the shape of the fruit of some varieties, which are white and resemble to chicken eggs shape. It is also called aubergine (French word) in Europe.It is being grown extensively in India, Bangladesh, Pakistan, China and the Philippines. Brinjal is known to be invaded

by many phytopathogens which are major constraints for the limited production and productivity of this crop. Among them, fruit rot caused by *Phomopsis vexans* is considered to be the most destructive disease of brinjal. Being a seed-borne pathogen, *P. vexans* establishes itself in seedlings and causes seedling death before it reaches maturity and also serves as a source of secondary inoculum. In the nursery, it causes damping-off of seedlings, and seedling blight and as the disease progress, an elongated blackish-to-brown lesion appears on the stem and branches. On leaves, it forms small circular, buff olive to cinnamon-buff spots with irregular blackish margins and on fruits, the disease appears as minute sunken greyish spots with a brownish halo, which later enlarge and produce concentric rings with yellow and brown zones (Panwar *et al.*, 1970). Reduction in germination, vigour and other seed quality parameters result in great economic loss, as it leads to poor production and productivity of the crop (Vishunavat and Kumar, 1993). Control of fruit rot by chemicals is easy but dependence on chemical methods of disease management may lead to residue and persistence problems, death of beneficial flora and fauna and evolution of fungicide-resistant pathogen population. The use of biocontrol agents to control plant diseases offers an excellent alternative to chemical control. In the present investigation, seed quality parameters of healthy and infected seeds of various genotypes of brinjal and their management using biocontrol agents were undertaken under laboratory conditions.

Materials and Methods

Raw material

Seeds from healthy and infected fruits ofseventeen brinjal genotypes were collected from the brinjal field of the Division of Vegetable Science, ICAR-Indian Agricultural Research Institute (ICAR - IARI), New Delhi, during 2017-2018 and revalidated in 2019-20 (Table 1). The seeds were extracted from the fermentation process by placing brinjal fruits in a container containing water for 24 hours and dried for a five hours under shade and both healthy and infected seeds were collected and maintained separately.

Seed Quality parameters

The mycelial growth on seeds was observed under the standard blotter paper method. The germination test was conducted according to International seed testing rules, 2018. The percentage of seed germination was recorded using four hundred seeds for each treatment by employing between the paper method with four replications. Ten normal seedlings were selected randomly from the germination test, shoot and root lengths of those selected seedlings were recorded for the calculation of seedling length. The mean value of seedling length was calculated and expressed in centimetres. For seedling dry weight, randomly taken ten normal seedlings were placed in butter paper and dried for 24 hours in a hot air oven maintained at 70 °C. The dry weight of the seedlings was recorded in an electronic balance and the average weight was computed and expressed in milligrams per ten seedlings. For electrical conductivity, 100 mg of seeds were soaked in 25 ml of distilled water for 24 hr at 20 °C. The seed leachate was collected by decanting and the electrical conductivity of the seed leachate was measured at room temperature with a conductivity bridge meter along with distilled water as control and was expressed as µS/cm/g (Dadlani and Agarwal, 1987). The vigour indices of seeds were calculated according to the method suggested by (Abdul-Baki and Anderson, 1973) and were expressed as a pure number.

Vigour Index (V I) I = Germination (%) x Total seedling length (cm)

Vigour Index (V I) II = Germination (%) x Seedling dry weight (mg)

Dual culture

In the dual culture technique, 15 ml of sterilized

Table 1. List of brinjal genotypes used for seed quality parameter studies.

G-181	G-204	G-17	PusaKaushal
PusaKranti	G-145	Pant Samrat	Pusa Uttam
Kushpada Local	G-23	Pusa Bindu	Arka Kusumakar
G-22	DB-9	Pusa Shymla	G-100 and G-40

potato dextrose agar was poured into sterile petri plates and allowed to solidify. The pathogen at one side of the petri plate and the antagonist organisms wereinoculated at the exact opposite side of the same plate by leaving gap of 3-4 cm gap. Each treatment was replicated three times. These plates were incubated at 25 ± 2 °C for seven days and colony diameter was recorded. Per cent inhibition over control was calculated as per the formula given by Vincent, (1947).

$$I = \frac{(C-T)}{C} \times 100$$

I = Percent inhibition of mycelium
C = Growth of mycelium in control
T = Growth of mycelium in treatment

Statistics

The analysis of variance was done using a completely randomized design analysis in the OPSTAT sheet. Statistical significance was tested using the "F" test. The critical difference was also used to test the difference between any two means.

Results and Discussion

Seed quality parameters

The germination percentage (mean value) of healthy seeds ranged between 75-83%, while infected seeds ranged between 3-56%. The reduction in germination percentage of diseased seeds varied between 14 to 67%. Maximum germination (83%) was recorded in healthy seeds of genotypes G-181 and G-204 which drastically fell down to 42% and 43% respectively in infected seeds (Fig. 1). Pan and Archarya, (1995) reported a decrease in the seed quality of different brinjal genotypes due to seed-borne infection by the pathogen.

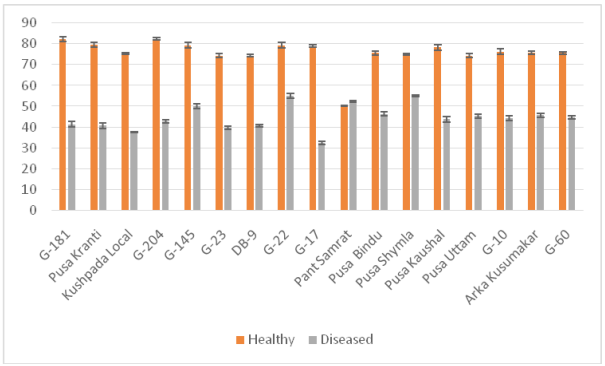

Fig. 1. Germination percentage (mean value in percentage) of healthy and infected seeds

The vigour index I of healthy seeds ranged between 1471-1628 cm, while infected seeds ranged between 16.80 cm to 647.6 cm. Maximum vigour of 1628 cm was recorded in healthy seeds of genotypes G-204 which drastically fell down to 381.6 cm in infected seeds (Fig. 2). The vigour index II of healthy seeds ranged between 1327-1614 mg, while infected seeds ranged between 33.75 mg to 305.0 mg. Maximum vigour of 1614 mg was recorded in healthy seeds of genotype G-181 which drastically fell down to 274.0 mg in infected seeds (Fig. 3).

The electrical conductivity of healthy seeds ranged between 38.31 to 35.62 µS cm^{-1}, while the infected seeds ranged between 70.12 to 62.23 µScm^{-1}. Minimum electrical conductivity of 37.92 was recorded in healthy seeds of genotypes G-181 which drastically raised to 70.12 µS cm^{-1} in infected seeds (Fig. 4). The seed quality of brinjal caused by *Phomopsis vexans* reduces the seed quality, vigour and viability which leads to an overall reduction in

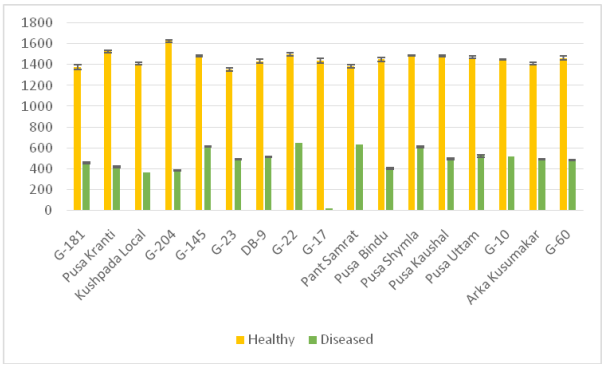

Fig. 2. Vigour index I (mean value in cm) of healthy and infected seed

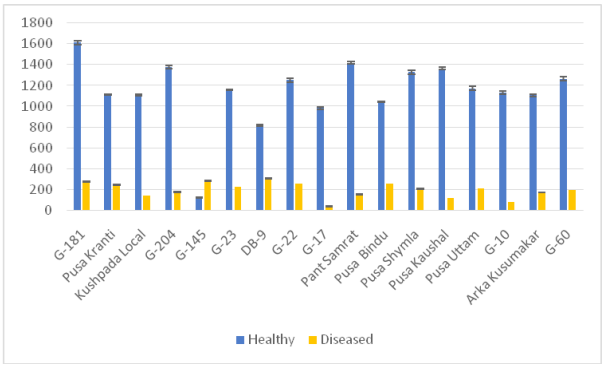

Fig. 3. Vigour index II (mean value in mg) of healthy and infected seeds

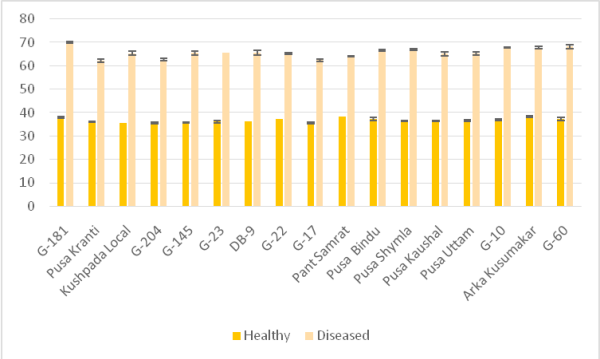

Fig 4. Electrical conductivity (mean value in μScm^{-1}) of healthy and infected seeds

the field establishment and plant population during crop production.

Dual culture

All the biocontrol agents under test were highly significantly over control in dual techniques against *Phomopsis vexans*. Among three bio-control agents evaluated *Trichoderma harzianum* showed maximum growth inhibition of the *Phomopsis vexans* which was 78.02%. Growth inhibition percentage of 38.46 % and 37.53 % was recorded with *Pseudomonas fluorescence* and *Bacillus subtilis* which is quite below compared to *Trichoderma harzianum* (Fig. 5). From the above, it is clear that the results of all the biocontrol tested by the dual culture method were effective against *Phomopsis vexans* and act as potential antagonistic agents. Among them, *Trichoderma harzianum* showed effective biocontrol against *Phomopsis vexans*. These findings were similar to work done by Jadeja (2003) who observed *Trichoderma harzianum* as effective biocontrol against *Phomopsis vexans*. At the natural condition *Trichoderma harzianum* has been considered as good model of biological control because of its ubiquitous nature, easy to isolate, rapid growth on many substrates affects wild range of plant pathogens acts as mycoparasite competes well for food and site, has enzyme system capable of attacking many plant pathogens and easy in application (Machenahalli *et al*., 2014). The mechanisms involved have been attributed to be mycoparasitim, antibiosis, competition for nutrients and space along with its ability to induce systemic resistance in the plants against the pathogens (Hermosa *et al*., 2012). This property has been further attributed due to the secretion of extracellular enzymes, including glucanases, chitinases etc., that degrade the pathogenic mycelia there by restricting its growth and further colonization in the host tissue (Singh *et al*., 2012).

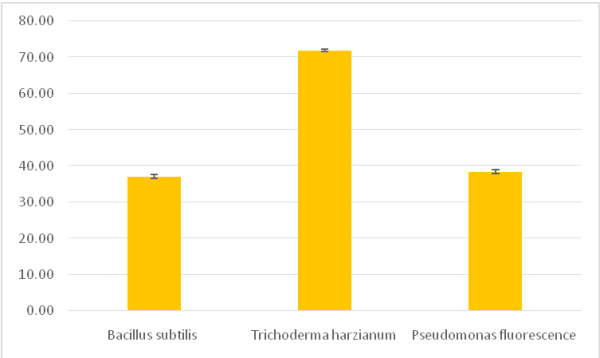

Fig 5. Percent inhibition (mean value) of *Phomopsis vexans* by different biocontrol agents

Conclusion

The use of biocontrol agents reduces the pathogen incidence thereby minimizing the impact of disease on yield and seed quality along with the minimal effect on environment flora and fauna. However, the activity of biocontrol may vary from place to place depending upon the experimental conditions. Therefore, validation under field conditions for its effect is necessary for giving recommendations to the farmers

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Conflict of Interest: On behalf of all authors, the corresponding author states that there is no conflict of interest.

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