World Journal of Pharmaceutical and Life Sciences
WJPLS
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SJIF Impact Factor: 6.129

TOXICOLOGICAL ASSESSMENT OF PSEUDOMONAS VIRIDIFLAVA ISOLATED FROM FABA BEAN CROP GROWN IN RAJASTHAN

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Article Received on 30/11/2023 **Article Revised on 20/12/2023** **Article Accepted on 10/01/2024**

ABSTRACT
Plant diseases showed the involvement of toxic metabolites, which plays a vital role in the disease development. These chemical metabolites of *Pseudomonas* bacterial pathogen may have severe effects on plants growth of Faba bean. Bacterial pathogen *Pseudomonas viridiflava* was isolated from symptomatic plant samples of faba bean crop of Rajasthan. Toxins isolated from bacterial cultural filterate at different intervals different periods i.e. 4, 16, 24, 30 hrs on seed embryos were used to estimate the toxic effects on seed viability and morphological growth. Faba Bean (*Vicia faba* L.) seed embryos completely lost their viability after 16 hrs treatment of bacterial cultural filtrates of *Pseudomonas viridiflava* gradually and unable to germinate.

KEYWORDS: Bacterial pathogen, Disease symptoms, Toxins, Cultural filterate, Faba bean.

INTRODUCTION

Phyto-pathogens mostly destruct their host plants by producing chemical substance such as toxins, which cause different symptoms including chlorosis, wilting, water soaked lesions necrosis, and finally the death of plants (Scheffer, 1983). Many phytotoxic secondary metabolites have been found associated with bacterial pathogens, which, causes symptoms in plants. Such phytotoxic metabolites include phaseolotoxin, tabtoxin and syringomycin and syringopeptin from *Pseudomonas syringae* pv *phaseolicola*. Toxins are chemical metabolites produced by bacterial pathogens which play a significant role in host (Plant) pathogen interactions. Toxins have very low molecular weight and produced by some pant pathogens which are capable to causing disease symptoms and infections in plants (Bilgrami, 1976) According to Amusa (1998) plant toxins are a chemical product of microbial pathogens, which often cause damage to plant parts like leaves, stem, flower and fruit and must be involved in disease development (Durbin, 2012). The pathogen and its toxic metabolite showed similar host specificity and be able to induce similar disease symptoms in plants. Phyto-toxins types-Various characteristics have been used for the classification of toxins that affect plants (Agrios, 2005). Some phytotoxins are classified as low molecular weight peptides, others have terpenoid structures and some contain carbohydrates (Amusa, 2000).

MATERIALS AND METHODS

The culture filterate of isolated bacterial pathogen (*Pseudomonas viridiflava*) was grown on nutrient agar broth to produce crude toxin (Fuchs, *et al.*, 1998). 50 ml of sterilized broth media was prepared and inoculated with 4 ml of the bacterial suspension and incubated at 25±2ºC for 30 days. The bacterial suspension was filtered and centrifuged for 25 min. at the speed of 10,000 rpm and the supernatant was used as crude bacterial toxins (Verma and Meena, 2021). The aqueous sol. of 2, 3, 5-triphenyl tetrazolium chloride (TTC) with a conc. of 0.1% was prepared in pH 7 of phosphate buffer for testing the viability of the embryo. The isolated embryos were treated with culture filtrate of isolated bacteria for different periods ie. 4,16, 24, 30 hrs. The control (untreated seeds) was treated with sterile distilled water. After the treatment for different time periods the data on germination of seeds in percentage and length of shoot and root in cm were recorded (Geng *et al*., 2012).

RESULTS AND DISCUSSION

After treating the healthy seeds with culture filtrate of bacterial isolate for 4, 16, 24, and 30 h, the morphological parameters such as root length, seed germination, and shoot length was measured to identify the toxic effects of isolated pathogen on faba bean growth. Treatment of *P. viridiflava* for 4h toxin the average shoot length of faba bean seedling was reduced. At 16h of treatment the shoot and root length was also found reduced. After 24 and 30h of treatment seed

viability was completely lost with culture filtrate of isolated pathogen. The maximum shoot length and root length was found 18.42 cm and 3.75 cm for control while it was 3.89 cm and 1.2 cm for *P. viridiflava* after 16h of treatment (Fig.1, Table1). So, the toxicological assessment concluded that the seeds viability is completely lost after infection of pathogenic bacterial strain (Sobiczewski, 2008 and Aktories, 2015). The virulent symptoms of *P. viridiflava* includes enzyme pectate lyase (Lipps and Samac, 2022). Therefore, bacterial pathogens and their toxins are the major source of significant problems in agriculture (Horbach *et al.*, 2011; Gallarato *et al.*, 2012).

Toxicological assessment of *Pseudomonas viridiflava* in Faba bean

(A) (B) (C) (D) (E)

Fig. 1: (A) Control (B) 4hrs (C) 16 hrs (D) 24 hrs (E) 30 hrs Toxin treatment with *P. viridiflava.*

Table 1: Effects of Toxins Produced by Bacterial Pathogen *Pseudomonas viridiflava*.

Isolated pathogen	Morphological parameters											
	4h			16h			24h			30h		
	Seed germination (%)	Shoot length (cm)	Root length (cm)	Seed germination (%)	Shoot length (cm)	Root length (cm)	Seed germination (%)	Shoot length (cm)	Root length (cm)	Seed germination (%)	Shoot length (cm)	Root length (cm)
Control (Untreated seeds)	100	18.42	3.75	100	18.43	3.73	100	18.41	3.71	100	18.39	3.72
Pseudomonas viridiflava	10	6	1.78	10	3.89	1.2	0	0	0	0	0	0

CONCLUSION

The results of toxicological assessment of toxin produced by *P. viridiflava* pathogen revealed that after 16h of treatment with *P. viridiflava* seed viability was completely lost and seed was unable to germinate. Phytotoxic results of experiment revealed that toxin produced by bacterial pathogen suppressed the seed viability and growth of Faba bean. However, no recorded data on toxicity of *Pseudomonas viridiflava* in Faba bean (*V. faba)* plant were previously available. Therefore, this study is one of the novel reports about the toxicity of *Pseudomonas viridiflava* on faba bean plant.

ACKNOWLEDGEMENT

I am highly thankful to Principal, HOD and faculty members of department of Botany, Raj Rishi Govt. College, Alwar for providing necessary infrastructure and support in many ways.

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