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“Transmission and Phyto-pathological effects of *Pseudomonas viridiflava* causing bacterial spot disease of *Vicia faba*”

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ABSTRACT: Legumes play an important nutritional role in the diet of people in developing countries. *Vicia faba* is an important legume crop, which has high protein content and efficient atmospheric nitrogen-fixing ability. Faba bean is a valued crop and provides nutritious food for an expanding world population and will become increasingly important with climate change. The nutritional value of faba bean in terms of nutrition and body health has been emphasized frequently by a many nutritionists. *P. viridiflava* has broad range of host and causes bacterial spot disease in *Vicia faba*. It shows a variety of pathogenic symptoms in plant parts such as leaves and stems. *P. viridiflava* inoculum can also come from contaminated seeds and reduce the crop yield. *P. viridiflava* is responsible to reduce the growth of crop and yield of faba bean.

KEYWORDS: *Vicia faba*, phyto-pathogen, transmission, bacteria, *P. viridiflava*.

I.INTRODUCTION

Legumes are the main source of protein and are well known for their nutritional value. They are relatively cheap source of protein. Leguminous crops such as lentils, peas, broad beans and soya beans are main components of a healthy diet (Nene, 1988). *Vicia faba* L., is important grain legume of family leguminosae. It has several common names such as Bakla, Kalamatar, Broad Bean, Horse Bean etc. The term ‘Fava’ bean is taken from the Italian word ‘Fava’ which means broad bean is extensively used in different countries. These are good source of protein, carbohydrate, dietary fiber, minerals and secondary metabolites such as phenolics, L-DOPA (Rabey *et al.*, 1992). The protein content of Faba bean is in the range of approximately 20-40% depending upon the seed quality of different varieties (Vidal-Valverde *et al.*, 1998).

Faba bean green pods and dry seeds used as vegetable and dal respectively. They also help to increase vital nitrogen in the soil (Etemadi *et al.*, 2018). Faba bean is infected by many pathogens. Some important fungal, bacterial and viral disease are Chocolate spot, Rust, Black root rot, Foot rots, Halo spot, Bacterial brown spot, Bean yellow mosaic virus etc. *Pseudomonas viridiflava* was first described by Burkholder in 1930 as a bean (*Phaseolus vulgaris*) pathogen. It is multi host pathogen. It can produce a variety of symptoms including necrosis, rots, blight and spots affecting different plant parts (Lipps *et al.*, 2022). It can cause bacterial blight in peas, rots in carrot, bacterial canker in stone fruits, bacterial leaf spot in pumpkin and soft rot in tomato and *Arabidopsis thaliana* (Goumas *et al.*, 1999). Typical symptoms infection in tomato are a general wilting and yellowing of the plants and brown black spots developing at the pruning sites of stem (Sarris *et al.*, 2012). In poppy infected plants were observed to turn brown to black streaks with translucent olive green edges at the base of stem and leaves. Whereas in pumpkin olive green, water soaked spots were observed which developed into pale brown lesions (Wilkie *et al.*, 1973). In peas pathogen affects the leaves, stipules and stems and showed water soaked, soft brown lesions.

In the present study infected seeds to seedling transmission and phyto-pathological effects of *Pseudomonas viridiflava* causing bacterial spot disease of faba bean were studied.

II. MATERIALS AND METHODS

The most common bacterial pathogen on faba beans are *Pseudomonas syringae*, *P. aeruginosa*, *Xanthomonas campestris* and *Pseudomonas viridiflava*. The infected plant debris or infected seeds of faba bean were the primary source of bacterial pathogen for new infection.

Faba bean growing fields were surveyed and infected plant samples were collected. Two types of seed samples of Faba bean carrying 80% and 89% natural infection of *Pseudomonas viridiflava* were selected for disease transmission studies. The experiment was done in triplicate forms. 30 seeds of per infected sample were placed on moist cotton blotters (10 Seed per plate) at equal distances.

To study phyto-pathological affect healthy faba bean seeds artificially treated with isolated bacterial broth culture were placed on moist cotton beds in petriplates and inoculated for 7 days at 25-30°C temperature. After that seedlings were again treated artificially with isolated bacterial broth culture by stabbing the cotyledons and performed blotter method and incubated 25-30°C for 7 days.

The percentage of seed germination, un-germinated seeds associated with pathogen, symptomatic seedling and mortality of seedlings were recorded timely.

In the pot experiment 30 infected seeds per category per sample were sown in plastic pots (5 seeds per pot). All the petriplate and pots were incubated at 25°C for 12-12 hour alternating cycles of light and dark photoperiod for 7 days. During the experimental studies percentage of seed germination, seedling symptoms and seedling mortality were recorded. There after bacterial colonies were isolated from infected seedling and seed to isolate pathogenic bacteria (Jain and Agarwal, 2011)

III. RESULT AND DISCUSSION

There are many seed borne bacteria, while a number of bacteria are serious pathogen on floweres and maturing seeds. These bacterial pathogen reduce the seed quality. The seed associated bacteria survive and proliferate by using nutrients and therefore, subsequently become pathogenic on seedlings and growing plants. Pathogenic bacteria proliferate in intercellular space after entering through stomata. These pathogens deliver some toxins into the plant cell to affect their fitness.

In the petriplate method 80% and 50% seed germination rate were recorded with 80% and 89% faba bean infected seeds in the present study respectively.

Some un-germinated seeds were shriveled and rolled out of these one seed of 89% infected seed sample is completely covered with white bacterial growth with rotting. Whereas the radicles of germinated seeds were also dried within the 7 days. Accordingly 20% and 50% mortality was observed with 80% and 89% infected seeds respectively.

In pot experiment 30% rate of seed germination was recorded with 80% faba bean infected seeds and 10% germination with 89% infected seeds. According to results, 70% and 90% mortality was observed with 80% and 89% infected faba bean seeds respectively. The non-germinated seeds showed shriveling and rotting. Due to heavy infection of the pathogen green and white bacterial growth on and around the bean seeds appeared. The infected radicle and plumule were used to isolate bacterial pathogen and identified using biochemical characteristics representative isolate of *Pseudomonas spp.* were identified as *Pseudomonas viridiflava* based on the determinative scheme as proposed by previous researchers (Lelliott et al., 1987).

In the phyto-pathological studies of artificially treated seed with *P. viridiflava* showed 60 percent germination while in control it was 100 percent. The length of shoot and root of control sample was 18.42cm and 3.75 cm respectively. Whereas radicle formation was not recorded in *P. viridiflava* infected seed sample.

The shoot and root length of artificially treated seeds raised seedlings with *P. viridiflava* was 1.10cm and 0.52cm respectively. Whereas the length of shoot and root of control seedling sample was 18.9cm and 3.88cm recorded respectively (Figure 1 and 2, Table 1).

The present study revealed that *Pseudomonas viridiflava* was found associated with infected faba beans. The bacterial pathogen was found transmitted with seed borne inoculum from seed to seedling to plant and causing bacterial spot in

the crop. The results of this study concluded that bacterial infection of plants reduce the seed viability and growth of beans.

Table 1: Phytopathological Effects and disease transmission of *Pseudomonas viridiflava* Isolated from Faba Bean

Parameters						
Sample	Control			<i>Pseudomonas viridiflava</i>		
	Seed germination (%)	Shoot length (cm)	Root length (cm)	Seed germination (%)	Shoot length (cm)	Root length (cm)
Seed treated	100	18.42	3.75	60	0.85	0
Seedling treated	100	18.9	3.88	60	1.10	0.52



(A)



(B)

Figure 1: Study of Phyto-pathological and transmission effects on seeds (A) Control (B) *Pseudomonas viridiflava*

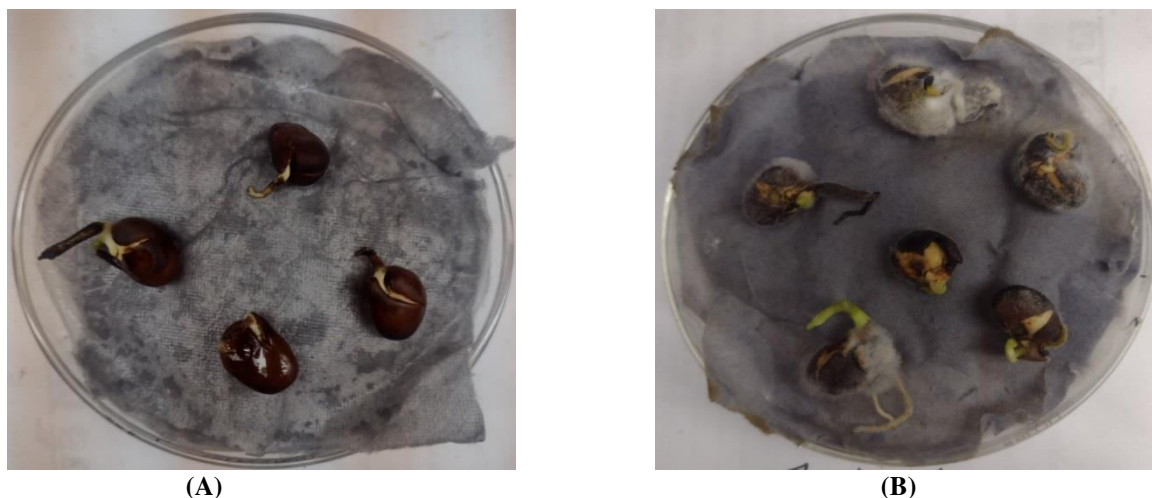


Figure 2: Study of Phyto-pathological and transmission effectson seedlings (A) Control (B) *Pseudomonas viridiflava*

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