



Effect of Age of Soybean Plants (*Glycine max* L.) on Susceptibility to Bacterial Blight caused by *Pseudomonas syringae* in Adamawa State

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Abstract

Soybean (*Glycine max*), a globally recognized crop plant has numerous industrial, domestic and nutritional values. However, its cultivation on the field has suffered lots of challenges especially infection by various micro-organisms such as fungi and bacteria. Soybean plants of three (3) and six (6) weeks old of the five different varieties were tested for susceptibility against the isolated strains of the bacteria (*Pseudomonas syringae*). The experiment was completely randomized with three replications. The data obtained were presented using bar charts and percentage error bars were used as the least significant difference (LSD). The result showed that there is great variation in susceptibility between 3-weeks and 6-weeks old Varieties of Soybean plants after seven days of being inoculated with Strains of *Pseudomonas syringae*. Three weeks old varieties of soybeans showed the highest susceptibility to Strains of *Pseudomonas syringae* at (243.6 mm) lesion size, while six weeks old varieties of soybeans inoculated with Strains of *Pseudomonas syringae* showed the least susceptibility at (153.2 mm) lesion size. It is recommended that a research work to be carried out on the effect of infection by pathogenic bacteria on the nutritional composition of soybeans seeds.

Keywords: Susceptibility, bacterial blight, soybean, Strains, lesion size.

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Introduction

Soybean (*Glycine max*) serves as a major human food and animal feed component due to its nutritional and health values (Alpha *et al.*, 2006). It is an important dietary source of protein, fat, fiber, minerals and vitamins, also cultivation of soybean occurs at different parts of Nigeria. This geographic distribution facilitates the spread of insect-pests and diseases (Al-Bari *et al.*, 2006). Hence, soybean is attacked by many different organisms, ranging from viruses, fungi, nematodes and bacteria (Ahmed, 2009). These pathogens can cause damage in seeds, roots, leaves, stems and pods, and usually are tissue-specific. The primary reason for this is the presence of wet and/or cool soil, which

enables the growth of pathogens (Al-Bari *et al.*, 2006). Plant protection in general and the protection of soybean against plant diseases in particular, have an obvious role to play in meeting the growing demand for food quality and quantity (Strange and Scott, 2005). Roughly, direct yield losses caused by pathogens, animals, and weeds, are altogether responsible for losses ranging between 20 and 40 % of global soybean productivity (Oerke, 2006). The crop losses due to pests and pathogens are direct, as well as indirect; they have a number of facets, some with short and others with long-term consequences (Alston *et al.*, 2009). Therefore about 40% adequately reflects the true costs of soybean losses to consumers, public

health, societies, environments, economic fabrics and farmers (Aubertot *et al.*, 2006). Bacterial infections are widespread diseases that occur mainly in the mid-to-upper and young leaves of the soybean plant (Alpha *et al.*, 2006). Soybean of different varieties are usually Susceptible to many pathogens (Sarkar *et al.*, 2015). Until recently, the Malayan variety was virtually the sole variety grown by farmers. This variety is low yielding, susceptible to bacterial diseases and is late maturing (Guha *et al.*, 2009). The aim of this research work is to determine the effect of age of soybean (*Glycine max* L.) on its susceptibility to *Pseudomonas syringae*.in Adamawa State

Materials and Methods

Susceptibility of Three and Six Weeks old Varieties of Soybean Plants (*in-vivo*) to the Four Identified Strains of Bacterial Pathogen (*Pseudomonas syringae*)

The five (5) most commonly cultivated soybean plants (TGX 1448-10F, TGX 1904-6F, TGX 1910-15F, TGX 1951-3F and TGX 1955-4F) varieties were grown in the screen house for three (3) and six (6) weeks respectively at the research farm of the department of plant sciences, Modibbo Adama University, Yola. The test for percentage susceptibility was carried out by obtaining a wire loop full of the pure culture of the strains of *Pseudomonas syringae* and dissolved in distilled water the mixture was then introduced to the surface of the leaves and stems of the healthy soybean plant using a sterilized spraying pump according to the method of (Aubertot *et al.*, 2006). The screen house was completely sealed to avoid contamination and kept at temperature 27±2 °C within the months of June to September, 2019. A similar set up was placed as control using sterile distilled water in place of the *Pseudomonas syringae* inoculant. It was kept and observed for seven (7) days to allow for possible symptoms development (Sarkar *et al.*, 2015). Data obtained was used to calculate and compare the percentage susceptibility for the two categories of the soybean plants varieties as well as cumulative susceptibility for all the five varieties. The formula is given by:

$$\%susceptibility = \frac{\text{Number of a variety of soybean plants infected}}{\text{Total number of a variety of soybean plants sowed}} \times 100$$

Bacterial Pathogenicity Test on Soybean plants

Healthy soybean plants of equal varieties were grown in the screen house located at Modibbo Adama University, Yola for three weeks. The test of pathogenicity (to determine the virulence of the pathogen) was carried out by obtaining a spatula full of the pure culture of *Pseudomonas syringae* and dissolved in distilled water, the mixture was then introduced to the surface of the leaves and stems of the healthy soybean plant using a sterilized spraying pump (Krawczyk, 2020). The screen house was completely sealed to avoid contamination. It was then kept at temperature 27±2 °C within the months of July to September, 2019. A similar set up was placed as control using sterile distilled water in place of the *pseudomonas syringae* inoculant. It was kept and observed for seven (7) days to allow for possible symptoms development and the isolates was re-isolated from the new host and shown to be the same as the originally inoculated pathogen. Data was taken every day for the period of seven (7) days. The reactions of the pathogen (Virulence) of the Bacterial blight on soybean plant was assessed and scored according to modified Emechebe's (1985) visual screen 0-5 in which:

- 0 no visible sign of infection
- 1 upto 20% of seedlings stems affected by bacterial blight
- 2 21 - 40% of seedlings stems affected by bacterial blight
- 3 41-60% of seedlings stems affected by bacterial blight
- 4 61-80% of seedlings stems affected by bacterial blight
- 5 More than 80% of seedlings stems affected by bacterial blight

The reactions of isolates based on the 0-5 visual scale was grouped in the following categories based on the Ratanacherdchail *et al.* (2010) pathogenic potential of grouping of isolates:

- 0-20% = low virulent group
- 21-40% = moderately virulent group
- 41-60% = high virulent group
- 61-80% = very high virulent group
- Above 80% = totally virulent group

Experimental Design and Data Analysis

The experiments was set up in a complete randomized design with three replication. The data obtained were presented using bar charts and percentage error bars were used as the least significant difference (LSD) at 5% probability level (Schaffer *et al.*, 2009).

Results

The result of soybean variety TGX 1448-10F at $p=0.05$ Figure 1 showed significant difference between 3 weeks and six weeks old. Mean differences exist in 3-weeks old, Strains (PsgCr12) showed the highest severity of (293 mm) lesion size while strain (PsgGr12) showed the least (255 mm) lesion size. Also differences was observed in 6-weeks old, Strains (PsgCr12) showed the highest severity of (203 mm) lesion size while strain (PsgBr6) showed the least (169 mm) lesion size.

The result in Figure 2 showed significant difference between 3 weeks and six weeks old TGX 1904-6F variety of soybean. 3-weeks old Variety showed Strains (PsgCr12) with the highest severity of (300 mm) lesion size while strain (PsgB110) showed the least (276 mm) lesion size. 6-weeks old Variety TGX 1904-6F of Soybean plants showed

Strains (PsgCr12) had the highest severity of (214 mm) lesion size while strain (PsgB110) showed the least (176 mm) lesion size.

Figure 3 shows significant difference between 3 weeks and six weeks old TGX 1910-15F variety of soybean. 3-weeks old showed strains (PsgCr12) had the highest severity of (270 mm) lesion size while strain (PsgGr12) showed the least (236 mm) lesion size. Also differences was observed in 6-weeks old Variety, strains (PsgCr12) showed the highest severity of (185 mm) lesion size while strain (PsgB110) showed the least (149 mm) lesion size.

There is significant difference between 3 weeks and six weeks old TGX 1951-3F variety of soybean in the result in Figure 4. Mean differences exists in 3-weeks old Variety. The Strains (PsgCr12) showed the highest severity of (242 mm) lesion size while strain (PsgGr12) showed the least (196 mm) lesion size. 6-weeks showed the highest severity of (136 mm) lesion size while strain (PsgB110) showed the least (99 mm) lesion size.

The result that there is significant difference between 3 weeks and six weeks old TGX 1955-4F variety of soybean is shown in Figure 5. The 3-weeks old showed that strains (PsgCr12) had the highest severity of (234 mm) lesion size while strain (PsgB110) had the least (128 mm) lesion size. 6-weeks old showed strains (PsgCr12) with the highest severity of (114mm) lesion size while strain (PsgB110) showed the least (93mm) lesion size.

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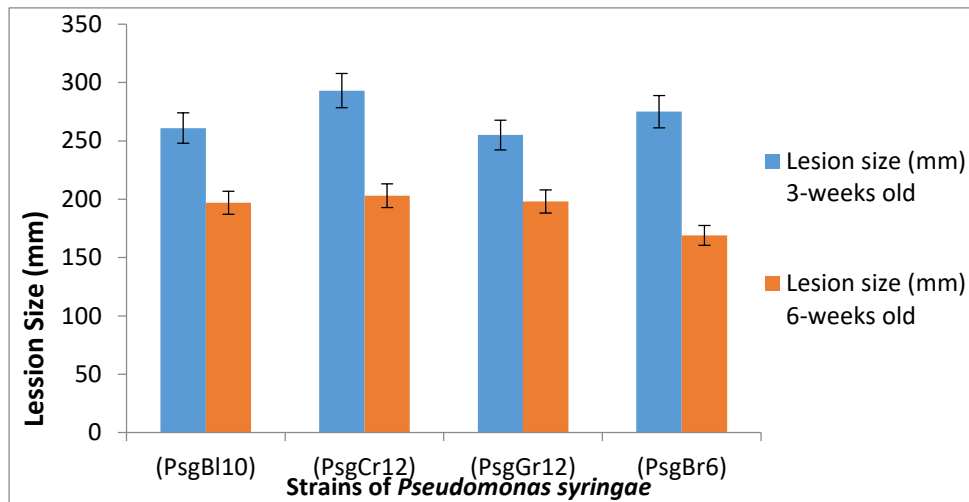


Figure 1: Susceptibility of Three and Six Weeks old Variety TGX 1448-10F of Soybean plants (mm) to the Four Identified Strains of *Pseudomonas syringae* (*in-vivo*)

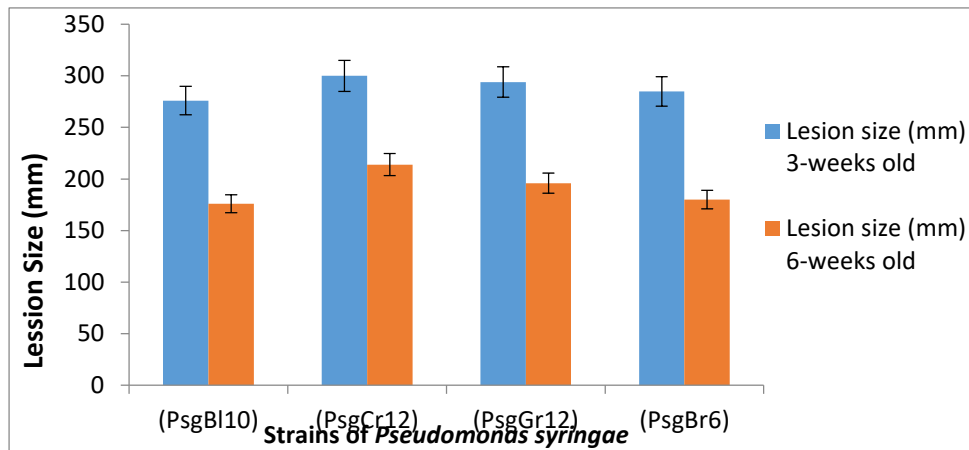


Figure 2; Susceptibility of Three and Six Weeks old Variety TGX 1904-6F of Soybean plants (mm) to the Four Identified Strains of *Pseudomonas syringae* (*in-vivo*)

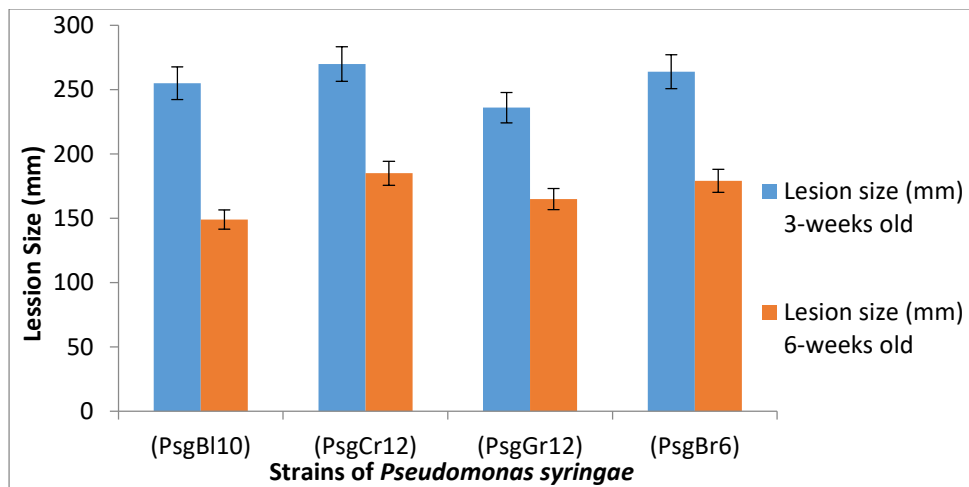


Figure 3; Susceptibility of Three and Six Weeks old Variety TGX 1910-15F of Soybean plants (mm) to the Four Identified Strains of *Pseudomonas syringae* (*in-vivo*)

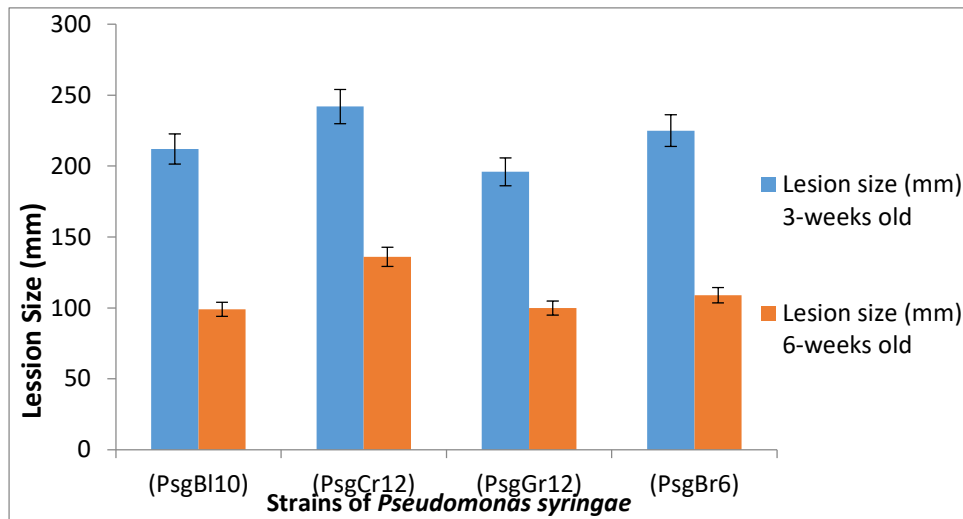


Figure 4; Susceptibility of Three and Six Weeks old Variety TGX 1951-3F of Soybean plants (mm) to the Four Identified Strains of *Pseudomonas syringae* (*in-vivo*)

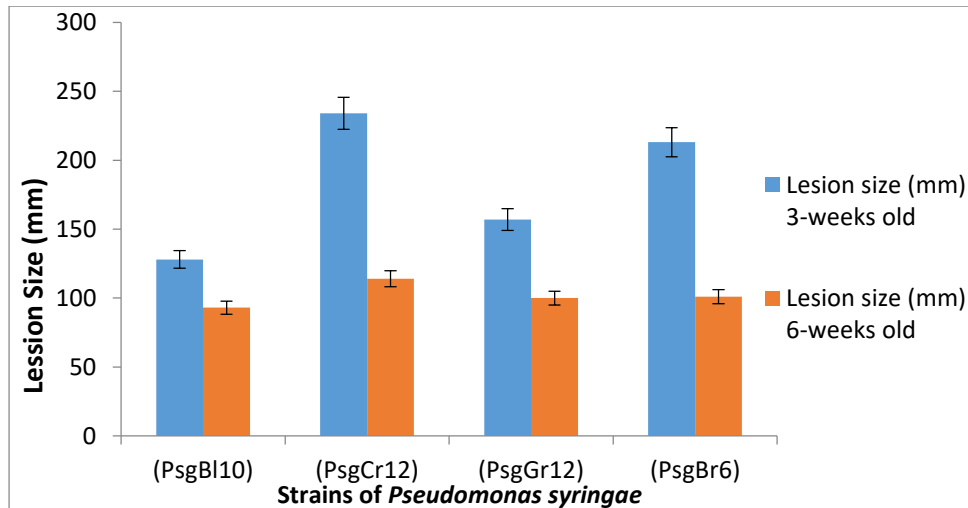


Figure 5; Susceptibility of Three and Six Weeks old Variety TGX 1955-4F of Soybean plants (mm) to the Four Identified Strains of *Pseudomonas syringae* (*in-vivo*)

The result in Figure 6 showed that there is significance difference between the three and six weeks old varieties of soybean inoculated with the strain of *Pseudomonas syringae*. There is difference in lesion size between 3-weeks and 6-weeks old Varieties of Soybean plants three weeks old varieties of soybeans showed the highest susceptibility to Strains of *Pseudomonas syringae* at (243.6mm) lesion size, while six weeks old varieties of soybeans inoculated with Strains of *Pseudomonas syringae* showed the least susceptibility at (153.2mm) lesion size.

Figure 7 showed the result of significant difference between three and six weeks old varieties (TGX 1448-10F, TGX 1904-6F, TGX 1910-15F, TGX 1951-3F and TGX 1955-4F) of soybean plants. There is difference in 3-weeks old among the five Varieties (TGX 1448-10F, TGX 1904-6F, TGX 1910-15F, TGX 1951-3F and TGX 1955-4F) of Soybean plants, TGX 1904-6F showed the highest susceptibility of (288.8 mm) lesion size while Variety TGX 1955-4F showed the least (183.0 mm) lesion size. There is difference in 6-weeks old among the five Varieties. However, there is no

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significant difference between Variety TGX 1448-10F and TGX 1904-6F, and both varieties showed the highest susceptibility of

(191.8 mm and 191.5 mm) lesion size respectively while Variety TGX 1955-4F showed the least (102.0 mm) lesion size.

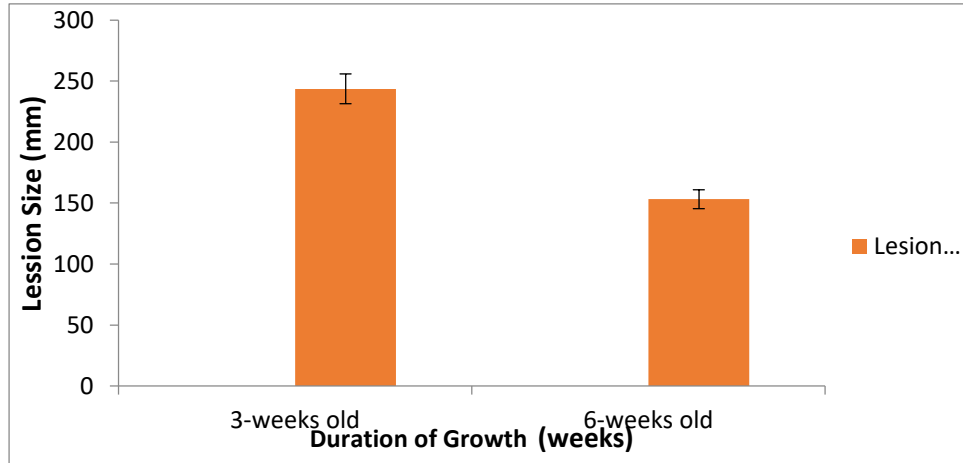


Figure 6; Susceptibility of Three and Six Weeks old Soybean plants to *Pseudomonas syringae* (*in-vivo*) (mm)

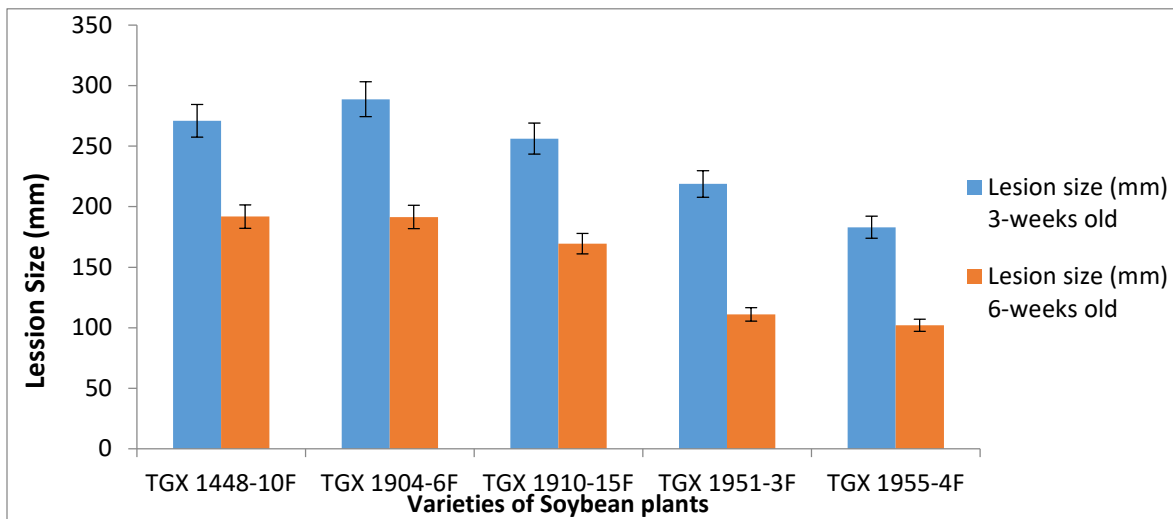


Figure 7: Susceptibility of Three and Six Weeks old Varieties (TGX 1448-10F, TGX 1904-6F, TGX 1910-15F, TGX 1951-3F and TGX 1955-4F) of Soybean plants to *Pseudomonas syringae* (*in-vivo*) (mm)

Discussion

Test for susceptibility among five different varieties of soybeans revealed there is great variation in lesion size between 3-weeks and 6-weeks old Varieties of Soybean plants after days of inoculation with Strains of *Pseudomonas syringae*. Three weeks old varieties of soybeans showed the highest susceptibility to strains of *Pseudomonas syringae* at (243.6mm) lesion size, while six weeks old varieties of soybeans inoculated

with Strains of *Pseudomonas syringae* showed the least susceptibility at (153.2mm) lesion size. El-shamy *et al.*, (2016) reported the potent powdery mildew resistance of cowpea, the mutant lines (mlo2-5, mlo6-2 and mlo12-1) was also previously found with respect to another adapted powdery mildew fungus on the tomato pathogen *O. neolycopersici*. Azeemi (2017) reported that transgenic soybean lines expressing part of the P3 and HC-pro genes showed a stable and

enhanced resistance to SMV-SC3, -SC7, -SC15, -SC18, and -R (a novel recombinant strain found in China) and have the potential to significantly increased soybean yield. With the continuous discoveries of defense mechanisms and the implementation of new molecular tools in breeding programs, generating efficient resistant plants will be faster to achieve (Gill *et al.*, 2015).

Among 3-weeks old, There is variation among the five Varieties of Soybean plants, variety TGX 1904-6F showed the highest susceptibility of (288.8 mm) lesion size while Variety TGX 1955-4F showed the least (183.0 mm) lesion size, while among 6-weeks old five Varieties of Soybean plants, there is no variation between Variety TGX 1448-10F and TGX 1904-6F, and both varieties showed the highest susceptibility of (191.8 mm and 191.5 mm) lesion size respectively while Variety TGX 1955-4F showed the least (102.0 mm) lesion size. The effect of SA levels on Arabidopsis resistance to insects can also be seen in the case of the *cpr1* and *cpr6* mutants, which exhibit constitutively elevated levels of SA. Larvae showed approximately 50% increase in weight gain on these mutants (Sarkar *et al.*, 2015). In addition, the triple mutant is also resistant to two non-adapted powdery mildew fungi (Oueslati *et al.*, 2020), the pea pathogen *Erysiphe pisi* and *Blumeria graminis* *F.sp. hordei* (Bgh), a pathogen of barley 11 (Sun *et al.*, 2015; Chakraborty and Bengal, 2012).

Conclusion

There is variation in susceptibility between 3-weeks and 6-weeks old Varieties of Soybean plants after seven days of being inoculated with Strains of *Pseudomonas syringae*. Three weeks old varieties of soybeans showed the highest susceptibility to Strains of *Pseudomonas syringae*, while six weeks old varieties of soybeans inoculated with Strains of *Pseudomonas syringae* showed the least susceptibility. There is the need for a research work to be carried out on the effect of infection by pathogenic bacteria on the nutritional composition of soybeans seeds.

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