

Silicon sources-controlled damping-off and enhanced health of pea (*Pisum sativum* L.)



Abeer A. ElGhanam, Mohamed M.H. Rahhal, Seham S. Ragab and Eman Y. Khafagi*¹

Address:

Plant Pathology Research Institute, Agricultural Research Center, Giza, Egypt

*Corresponding author: **Eman Y. Khafagi**, email: dr_emankhafagi@yahoo.com

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ABSTRACT

This study evaluated the response of three pea cultivars (*Pisum sativum* L.): Master B, Blmewar, and Sugar Gum to damping-off under greenhouse conditions. It also investigated the effect of seed soaking in silicate solutions on damping-off and plant growth under field conditions. Fungal pathogens identified were *Fusarium oxysporum*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, and *Pythium ultimum*. These pathogens caused varying levels of damping-off in pot experiments. Field trials at the Etay El-Baroud Agricultural Research Station, El-Beheira Governorate, were conducted over two seasons (2021/2022 and 2022/2023) using magnesium silicate, potassium silicate, and Rizolex/Thiram treatments. Sugar Gum showed the lowest damping-off rates and superior growth characteristics, including root length, fresh and dry weight, and seed protein content. It also had the lowest polygalacturonase and Cx-cellulase activities. Blmewar had the highest seed weight per plant. Rizolex/Thiram was the most effective treatment, but both silicates also significantly reduced damping-off compared to the control. MgSiO₃ (4 g/L) was particularly effective, showing the lowest pre-emergence damping-off and improved growth parameters.

Keywords: Pea, magnesium silicate, potassium silicate, damping-off stress, lignin, nitrogenase, bacterial nodules , polygalacturonase, Cx-cellulase

INTRODUCTION

Pisum sativum L., or pea, is a significant global legume crop, renowned for its rich nutritional content, including proteins, carbohydrates, dietary fiber, vitamins, and minerals. Recent research highlights the presence of macronutrients and bioactive compounds such as polyphenols, flavonoids, and phenolic acids in peas, which contribute to health benefits like antioxidant, anti-inflammatory, and antimicrobial effects. Additionally, peas have a low glycemic index, beneficial for managing type 2 diabetes and reducing blood cholesterol levels (Wu *et al.*, 2023). Peas are highly vulnerable to pre-emergence damping off and post-emergence root and foot rots caused by soil and seed-borne fungi, significantly limiting their production. The use of chemical fungicides, while effective, raises concerns due to their toxic effects on non-target organisms, environmental contamination, and the emergence of resistant pathogen strains (Lamichhane *et al.*, 2017).

Recent studies stress the importance of integrated pest management practices and developing resistant pea varieties through advanced breeding techniques to address these challenges while promoting sustainable agriculture (Zhou *et al.*, 2023). Although silicon (Si) is not officially classified as an essential nutrient for plant growth, extensive research has demonstrated its beneficial effects on various growth parameters across multiple crops, leading to significant yield increases (Mir *et al.*, 2022). Studies have shown that silicate enhances plant resistance against a wide range of fungal, bacterial, and viral diseases in different plant species through physical, biochemical, and molecular mechanisms (Rodrigues and Datnoff 2015; Islam *et al.*, 2020). These mechanisms include cell wall stiffening, papillae formation, callose deposition, signal transduction, and the induction of stress-responsive gene expression (Wang *et al.*, 2017; Gulzar *et al.*, 2021).

The application of silicon has proven to be an effective method for controlling various soil-borne fungal diseases. It has been shown to mitigate the effects of *Fusarium oxysporum* f. sp. *radicis-lycopersici*, which causes crown and root rot in tomato seedlings (Huang *et al.*, (2011); *Fusarium oxysporum* f. sp. *vasinfectum* in cotton (Whan *et al.*, 2016); root rot caused by *Rhizoctonia solani* and *Fusarium solani* in strawberries and white rot caused by *Stromatinia cepivora* in onions and garlic (Elshahawy *et al.*, 2021). This study aims to optimize silicon application methods, study fungal pathogenicity on pea cultivars, assess silicate sources' effectiveness in reducing damping-off disease, and evaluate its impact on legume growth and symbiosis. It aims to integrate silicon-based treatments for sustainable agricultural practices, enhancing disease resistance and crop productivity.

MATERIALS AND METHODS

Study Location, Duration and source of Cultivars:

The study was conducted at the Etay El-Baroud Agricultural Research Station during the 2021/2022 and 2022/2023 seasons. Cultivars obtained from Veg. Res. Dept., Hort. Res. Inst., Agric. Res. Center, Giza, Egypt

Isolation and identification of the associated fungi:

Pea plants with damping-off and root rot symptoms were collected from El-Behera governorate, Egypt. Fungi were isolated on PDA medium and purified using hyphal tip and single spore techniques (Dhingra and Sinclair, 1995). Identification was based on cultural, morphological, and microscopic characteristics according to (Booth (1971); (Van der Plaats-Niterink, (1981) and Barnett, and Hunter (1986). Pure cultures were stored on PDA slants at $4\pm 1^\circ\text{C}$.

Preparation of inoculum:

Fungi were grown on PDA for 7 days at 25°C . Mycelium discs (two discs 5 mm) were transferred to cornmeal sand medium and incubated at 28°C for 15 days. according to (Muhanna *et al.*, 2018).

Pathogenicity test in greenhouse:

Seeds of Master B, Blmewar, and Sugar Gum cultivars were used. Soil was infested with fungal inoculum at 3% (w/w) according to (Habashy *et al.*, 2016) and sown with five seeds per pot, with three replicates for each treatment. Control pots contained sterilized soil mixed with autoclaved cornmeal medium. Pots were distributed under the greenhouse conditions in complete randomized design.

Disease assessment:

The percentage of pre- and post-emergence damping-off as well as surviving plants in each treatment were determined 15,30 and 45 days after sowing, respectively using the formula mentioned by (El-Helaly *et al.*, 1970).

Field experiments:

Field experiments used a split-plot design with three pea cultivars and six treatments mentioned in Table (1) in the sub-plots. Each plot was 12 m^2 with 5 ridges each of 0.6 m width and 4.0 m length. Soil irrigation was performed one week before sowing. Seeds were soaked in solutions of silicate salts and fungicide for 12 h, then sown 10 cm apart. Sowing occurred on October 15th for both seasons. Standard farming practices were followed.

Table 1. Soaking treatments

Abbreviation	Soaking treatments
MgSiO ₃ 2g/L	Magnesium silicate 2 g/L
MgSiO ₃ 4g/L	Magnesium silicate 4 g/L
K ₂ SiO ₃ 2g/L	Potassium silicate 2 g/L
K ₂ SiO ₃ 4g/L	Potassium silicate 4 g/L
R/T	Rhizolex / Thiram 3 g/L (positive control)
Water	Negative control

Silicate salts:

Magnesium silicate (MgSiO₃) and potassium silicate (K₂SiO₃) were obtained from El Gomhoria Company for chemicals, Cairo, Egypt.

Fungicide:

Rhizolex / Thiram (Thiram + Tolclofos-methyl Model: 30% + 30% WP)
Chemical Abstracts name: O-(2,6-dichloro-4-methylphenyl) O,O-dimethyl phosphorothioate

Growth Parameters:

Three plants were selected randomly from each replicate unit to measure the vegetative growth characteristics:

- Root fresh weight /plant (g) were estimated as the average of three plants chosen randomly after 65 days from sowing in the first and second seasons.
- Root dry weight /plant (g) were estimated after drying in an electric oven at 70°C for 48 h, till constant dry weight.
- Root length (cm).
- Number of bacterial nodules/roots.

Seed Weight:

At harvesting time (after 65 from sowing in the first and second seasons, respectively) 3 plants were randomly taken from each replicate to determine seed weight/plant (g).

Biochemical Changes Associated with Infection:

Determination of Protein Content in Pea Seeds:

The Protein determination in seed (g/kg-1seed) using the Kjeldahl method after harvest according to (Purificacion *et al.*, 2012).

Determination of leaf chlorophyll:

Total chlorophyll content was measured at 50 days after sowing date (flowering and pod set stage) in fresh leaves determined by using Minolta chlorophyll meter SPAD – 501 as SPAD units (Yadava, 1986). Total soluble solids (T. S. S. %) in fresh seeds was determined by using a hand Refractometer according to (A.O.A.C., 1990).

Preparation of enzyme extract:

After 50 days of cultivation one-gram roots of each treatment was freezed then blended in 10 ml 0.1 M phosphate buffer (pH 7.0) in a previously chilled. The homogenate was then centrifuged at 10,000 rpm for 20 min in a refrigerated centrifuge at 0–4°C. The supernatant obtained was referred to as crude extract and stored in freezer for enzyme assays.

Polygalacturonase assessment:

Polygalacturonase (PG) activity was determined by measuring loss in viscosity of the reaction mixtures according to the method of (Muhanna *et al.*, 2018).

Cellulase assessments:

Cellulase (Cx) activity was determined in terms of loss of viscosity (%) using the following formula (Muhanna *et al.*, 2018).

Estimation of nitrogenase using a colorimetric determination for ethylene:

The assay is applied to estimation of nitrogenase in nodulated pea roots by measuring the ethylene produced from acetylene according to (Vessey, 1994).

Statistical analysis:

Data obtained were subjected to the analyses of variance for split-plot design followed by compared means with L.S.D at the level probability 5%. ANOVA was performed for the experiment using CoStat© Ver. 6.400 statistical data analytical software.

RESULTS

Isolation and identification of the associated fungi

Isolation of fungal pathogens from diseased root pea samples collected from the experimental farm of Etay El-Baroud Agricultural Research Station, El-Behera governorate are presented in Table (1). The isolated fungi were identified, and their frequency percentages were calculated. *Fusarium oxysporum* showed the highest percentage followed by *Rhizoctonia solani* Kühn, *Sclerotinia sclerotiorum* (Lib.) and *Pythium ultimum* with averages of 35.25, 25.89, 18.06 and 15.09%, respectively at the same time a low frequency of minor other fungi (5.11%) was calculated.

1. Pots experiment

Results in Table 2 indicated that all the tested fungi caused pre and post emergence damping off in the tested cultivars *i.e.*, Master B, Blmewar and Sugar gum. In case of the soil inoculated with *Fusarium oxysporum*, the highest percentage of pre - emergence damping off was

Table 1. Frequency percentage of fungi isolated from the rotten roots and wilted pea plants.

Isolate fungi	Frequency (%)
<i>Fusarium oxysporum</i>	35.25
<i>Rhizoctonia solani</i>	25.89
<i>Sclerotinia sclerotiorum</i>	18.66
<i>Pythium ultimum</i>	15.09
Other fungi (neglected)	5.11

cleared with Master B cv (60.00%) followed by Blmewar and Sugar gum being 46.67 and 40.00 %, respectively. Meanwhile in the case of *Rhizoctonia solani* the highest percentage of pre - emergence damping off was recorded Master B with average of 53.33 % followed Blmewar and Sugar gum with the same average of 40.00 %. In respect to *Sclerotinia sclerotiorum*, Master B also had the highest value of pre- emergence being 46.67 % followed by Blmewar and Sugar gum being 26.67 and 20.00 %, respectively. Finally, the same trend cleared in case of *Pythium ultimum* where Master B had the highest value with 40.00 % followed by Blmewar and Sugar gum with the same value of 20.00 %, respectively. On the other hand, the cultivars arrangement differed in case of the post-emergence caused by *Fusarium oxysporum* where Blmewar had the highest value being 33.33 % followed by Master B and Sugar gum being the same value (20.00 %). Regarding *Rhizoctonia solani*, both cultivars; Blmewar and Sugar gum had the same value with 33.33 % followed by Master B with 20.00 %. In case of *Sclerotinia sclerotiorum*, both cultivars Master B and Blmewar had the highest values being 26.67 % followed by Sugar gum being 20.00 %. Finally Master B had the

Table 2. Effect of the tested fungi on disease incidence of damping-off with three pea cultivars under greenhouse conditions.

Cultivar		Pre-emergence %	Post-emergence %	Survival plants %
<i>Fusarium oxysporum</i>				
Master B		60.00 a	20.00 b	20.00 e
Blmewar		46.67 b	33.33 a	20.00 e
Sugar gum		40.00 c	20.00 b	40.00 d
Control	Master B	20.00 d	6.67 c	73.33 c
	Blmewar	6.67 e	6.67 c	86.67 b
	Sugar gum	0.00 f	0.00 d	100.00 a
LSD 5%		5.09	3.85	8.11
<i>Rhizoctonia solani</i>				
Master B		53.33 a	20.00 b	26.67 d
Blmewar		40.00 b	33.33 a	26.67 d
Sugar gum		40.00 b	33.33 a	26.67 d
Control	Master B	20.00 c	6.67 c	73.33 c
	Blmewar	6.67 d	6.67 c	86.66 b
	Sugar gum	0.00 e	0.00 d	100.00 a
LSD 5%		3.47	3.62	7.49
<i>Sclerotinia sclerotiorum</i>				
Master B		46.67 a	26.67 a	26.66 f
Blmewar		26.67 b	26.67 a	46.66 e
Sugar gum		20.00 c	20.00 b	60.00 d
Control	Master B	20.00 d	6.67 c	73.33 c
	Blmewar	6.67 e	6.67 c	86.66 b
	Sugar gum	0.00 f	0.00 d	100.00 a
LSD 5%		6.14	1.92	10.25
<i>Pythium ultimum</i>				
Master B		40.00 a	26.67 a	33.33 e
Blmewar		20.00 b	20.00 b	60.00 d
Sugar gum		20.00 b	20.00 b	60.00 d
Control	Master B	20.00 c	6.67 c	73.33 c
	Blmewar	6.67 d	6.67 c	86.66 b
	Sugar gum	0.00 e	0.00 d	100.00 a
LSD 5%		5.77	1.92	9.82

highest value with 26.67 % followed by Blmewar and Sugar gum with the same value of 20.00 %. In case of the survival plants %, its values increased with the cultivars had the least damping-off vales as it clear with Sugar gum cultivated in the soil infested with *Sclerotinia sclerotiorum* and Blmewar and Sugar gum cultivated in the soil infested with *Pythium ultimum* with the same value of 60.00 %. In contrast, the least survival plants % cleared with both Master B and Blmawer with average of 20.00 %. Statistically, the differences between the above values were significant.

2. Field experiments:

2.1. Effect of seed soaking treatments on cultivars and their interaction for the incidence of pea damping off disease under field conditions:

In these experiments, efficacy of pea seed soaking in magnesium silicate ($MgSiO_3$), potassium silicate (K_2SiO_3) at 2 and 4 g/L and the chemical fungicide; Rizolex/Thiram (R/T 3g/L) on damping off incidence and survived plants of pea under field conditions were studied. Results in Table (3) exhibited that Sugar gum cultivar had the least pre-emergence at the two seasons with averages of 23.32 and 23.64% followed by Blmewar and Master B, respectively. The statistical analysis showed that the differences between the values of sugar gum cultivar and both of Master B and Blmewar cultivars are significant. In case of post-emergence, Blmewar and Master B had the least values in the first and second seasons with averages of 11.24 and 12.38%, respectively. Soaking treatments significantly decreased pre-emergence damping-off compared to untreated control. The least percentages were obtained from treatments of R/T (3g/L), $MgSiO_3$ (4g/L) and K_2SiO_3 (4g/L) with averages of 5.17, 20.73 and 32.90% for the first season and 10.33, 24.99 and 35.17% for the second season, respectively. For the interaction effect, soaking seeds for each cultivar with R/T (3g/L) had the least pre-emergence damping-off with light variations in its arrangements for both seasons. Sugar gum x $MgSiO_3$ (4g/L) had considerable decrease in pre-emergence for both seasons. Also, the same trend cleared up in case of post-emergence damping-off where R/T had the least value followed by $MgSiO_3$ (4g/L), and K_2SiO_3 (4g/L) in the first season with averages of 4.24, 10.95 and 11.22%, respectively. In this respect, R/T (3g/L) had

the least value followed by K_2SiO_3 (4g/L) and $MgSiO_3$ (4g/L) with averages of 4.53, 9.71 and 11.87%, respectively in the second season. Also, results showed that all the tested treatments significantly increased the percentage of surviving plants compared with the control. For the interaction effect, soaking seeds for each cultivar with R/T (3g/L) had the least pre- and post – emergence damping-off with light variations in its arrangements for both seasons. Sugar gum x $MgSiO_3$ (4g/L) had considerable decrease in pre- emergence for both seasons.

In respect to post-emergence damping-off, Blmewar x $MgSiO_3$ (4g/L) and Blmewar x K_2SiO_3 (4g/L) had noticeable decrease for the first and second seasons, respectively.

Table3. Effect of pea cultivars, seed soaking treatments and their interaction on pre -, post - emergence damping - off and survival % during two successive seasons: 2021/2022 and 2022/2023 under field conditions.

Parameter	Pre-emergence damping-off %				Post-emergence damping-off %				Survival plants %			
	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean
Treatment	1 st season											
$MgSiO_3$ 2g/L	45.47	36.60	37.80	39.96 B	12.65	10.54	14.57	12.59 B	34.35	58.69	68.70	53.91 CD
$MgSiO_3$ 4g/L	18.90	30.00	13.30	20.73 D	11.29	8.33	13.23	10.95 C	68.83	54.83	78.99	67.55 B
K_2SiO_3 2g/L	52.20	37.70	22.20	37.37 B	13.78	11.57	12.87	12.74 B	34.02	42.82	64.93	47.26 D
K_2SiO_3 4g/L	36.60	46.60	15.50	32.90 C	13.94	10.53	9.20	11.22 BC	49.43	51.70	75.30	58.81 C
R/T 3g/L	6.70	4.40	4.40	5.17 E	3.57	3.43	5.78	4.26 D	89.77	92.13	89.78	90.56 A
Control	62.20	59.90	46.70	56.27 A	20.45	23.05	21.00	21.50 A	26.51	31.67	38.77	32.32 E
C. mean	37.01 A	35.87 A	23.32 B		12.61	11.24	12.78		50.49 B	55.31 B	69.41 A	
LSD 5% C	2.27				1.75 (NS)				7.82			
T	2.46				1.62				6.65			
C x T	5.98				1.89				7.74			
	2 nd season											
$MgSiO_3$ 2g/L	48.90	39.90	22.19	37.00 C	13.45	11.10	17.05	13.87 B	29.55	53.67	64.77	49.33 BCD
$MgSiO_3$ 4g/L	25.50	34.00	15.45	24.99 D	11.19	8.84	15.59	11.87 BC	61.01	48.93	60.75	56.90 B
K_2SiO_3 2g/L	58.90	49.90	27.71	45.50 B	13.57	11.29	11.00	11.95 BC	30.46	38.75	61.53	43.58 CD
K_2SiO_3 4g/L	43.30	43.30	18.91	35.17 C	10.56	7.82	10.74	9.71 C	43.17	48.88	70.13	54.06 BC
R/T 3g/L	15.50	7.80	7.70	10.33 E	3.95	3.61	6.04	4.53 D	83.55	88.61	86.23	86.13 A
Control	65.50	62.20	49.89	59.20 A	21.58	33.60	23.73	26.30 A	23.31	28.96	34.44	35.57 D
C. mean	42.93 A	39.52 A	23.64 B		12.38	12.71	14.62 A		45.18 C	51.30 B	66.31 A	
LSD 5% C	6.44				2.19 (NS)				6.54			
T	3.62				3.31				14.39			
C x T	4.21				3.85				16.74			

2.2. Effect of seed soaking treatments on cultivars and their interaction for some pea growth characters:

2.2. a. root length, root fresh weight and root dry weight:

Data in Table (4) appear that Sugar gum had the highest values of root length (cm), root fresh and dry weight (g) in both seasons followed by Master B and Blmewar, respectively. The results showed that soaking treatments significantly increased the previous parameters compared to untreated control treatment. Statistically the differences between silicate treatments are significant. Although R/T treatment had the highest values for the above parameters, soaking pea seeds in $MgSiO_3$ (4g/L), $MgSiO_3$ (2g/L) and K_2SiO_3 (4g/L) had noticeable altitude values. $MgSiO_3$ (4g/L) had 10.267 cm, 12.19 g and 4.42 g for root length, root fresh and dry weight followed by K_2SiO_3 (4g/L) and $MgSiO_3$ (2g/L) in the first season. At the second season the obtained results were closed to that of the first season.

Table 4. Effect of pea cultivars, seed soaking treatments and their interaction on root length (cm), root fresh and dry weight (g)/plant during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

Parameter	Root length (cm)				Root fresh weight (g)				Root dry weight (g)			
	Master B	Blmewar	Sugargum	T.mean	Master B	Blmewar	Sugargum	T.mean	Master B	Blmewar	Sugargum	T.mean
Treatment	1 st season											
MgSiO ₃ 2g/L	8.40	4.67	11.07	8.04 B	11.34	4.14	15.20	10.20 C	2.83	2.36	3.65	2.95 C
MgSiO ₃ 4g/L	9.10	7.83	13.87	10.27 A	12.86	4.76	18.94	12.19 B	4.20	3.55	5.50	4.42 B
K ₂ SiO ₃ 2g/L	5.00	3.90	8.03	5.64 C	9.83	3.22	10.81	7.95 D	2.31	1.54	2.54	2.13 D
K ₂ SiO ₃ 4g/L	7.37	5.43	10.03	7.61 B	10.30	3.64	13.99	9.31 CD	3.14	2.69	3.30	3.04 C
R/T 3g/L	10.33	8.53	14.70	11.19 A	17.78	5.35	27.96	17.03 A	4.43	3.45	6.75	4.88 A
Control	2.67	2.00	5.70	3.46 D	3.79	3.27	5.07	4.04 E	1.09	0.91	1.84	1.28 E
C. mean	7.14 B	5.39	10.57 A		10.98 B	4.06 C	15.33 A		3.00	2.42	3.93	
LSD 5% C	2.15 (NS)				4.31				2.19 (NS)			
T	1.19				1.68				0.13			
C x T	1.39				1.95				0.15			
	2 nd season											
MgSiO ₃ 2g/L	9.00	5.80	14.07	9.62 C	10.85	3.80	13.37	9.34 C	2.30	2.02	2.81	2.38 D
MgSiO ₃ 4g/L	10.27	8.77	15.40	11.48 B	11.67	4.27	18.37	11.44 B	3.57	3.57	4.43	3.63 B
K ₂ SiO ₃ 2g/L	6.28	4.83	9.53	6.88 D	9.20	2.83	10.36	7.47 D	1.77	1.77	2.24	1.79 E
K ₂ SiO ₃ 4g/L	8.23	6.23	12.20	8.89 C	10.84	3.67	13.69	9.40 C	2.80	2.80	2.88	2.62 C
R/T 3g/L	11.83	10.50	17.83	13.39 A	13.48	4.80	23.67	13.98 A	3.77	3.77	5.80	4.19 A
Control	6.23	4.10	7.30	5.88 D	2.80	2.20	8.28	4.43 E	0.89	0.79	1.02	0.90 E
C. mean	8.64 B	6.71 B	12.72 A		9.81 B	3.60 C	14.62 A		2.51 BC	2.04	3.20 A	
LSD 5% C	2.43				3.21				0.22			
T	1.57				1.28				0.12			
C x T	1.83				1.49				0.13			

2.2. b. seed weight/plant (g):

Data in Table (5) exhibited that Blmewar cv. had the highest seed yield/plant in both seasons with averages of 47.78 and 45.19 g. Also, data showed that R/T (3g/L) treatment had the highest values for both seasons. In this respect MgSiO₃ (4 and 2 g/L) had considerable increase compared to K₂SiO₃ (2 and 4 g/L). The differences between soaking treatments were significant in both seasons. In concern to the interaction, the statistical analysis showed that the differences between the values of seed weight/plant (g) are significant. Generally soaking Blmewar seeds in the solutions of MgSiO₃ or K₂SiO₃ (2 and 4g/L) led to noticeable increase in seed yield/plant compared to Master B and Sugar gum.

2.2.c. number of root bacterial nodules/plant, nitrogenase activity and protein content g kg⁻¹ seed:

Data in Table (6) appear that Sugar gum cv. had the highest number of bacterial nodules/plants for both seasons with averages of 8.06 and 7.06, seed protein content with averages of 110.95 and 111.77 g kg⁻¹ seed and nitrogenase activity in the first season only with an average of 0.46.

Table 5. Effect of seed pea soaking treatments, cultivars and their interaction on seed weight/plant during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

seed weight/plant				
Cultivar	Master B	Blmewar	Sugar gum	Treatment mean
Treatment	1st season			
Mg SiO ₃ 2g/L	22.71	56.00	16.87	31.86 B
Mg SiO ₃ 4g/L	23.69	56.92	17.93	32.85 B
K ₂ SiO ₃ 2g/L	14.09	36.80	10.81	20.57 D
K ₂ SiO ₃ 4g/L	14.63	45.72	12.97	24.44 C
T/R 3g/L	27.04	62.17	18.18	35.80 A
Control	10.99	29.07	8.32	16.13 D
Cultivar mean	18.86 B	47.78 A	14.18 B	
LSD 5% C	5.18			
T	3.86			
C x T	4.49			
	2nd season			
Mg SiO ₃ 2g/L	22.04	55.00	16.28	31.11 A
Mg SiO ₃ 4g/L	22.80	56.62	17.34	32.25 A
K ₂ SiO ₃ 2g/L	13.44	35.81	10.21	19.82 BC
K ₂ SiO ₃ 4g/L	14.51	41.12	10.90	22.18 B
T/R 3g/L	26.61	53.52	18.38	32.84 A
Control	10.64	29.06	9.62	16.44 C
Cultivar mean	18.34 B	45.19 A	13.79 C	
LSD 5% C	4.36			
T	4.32			
C x T	5.03			

In respect to seed soaking treatments, it can be noticed that all treatments significantly increased the number of bacterial nodules/plants, nitrogen activity and seed protein content compared to untreated control. R/T (3g/L) had the highest values for both seasons for number of bacterial nodules/plant and seed protein content with averages of 10.78 and 112.50 g kg⁻¹ followed by MgSiO₃ (4g/L) (9.33 and 112.27 g kg⁻¹seed), respectively. In case of nitrogenase activity, MgSiO₃ (4g/L) had the highest value followed by R/T (3g/L) in the first season with averages of 0.56 and 0.51, respectively, but in the second season R/T (3g/L) had the first grade followed by MgSiO₃ (2g/L) but the differences are not significant. From the interaction, there are significant differences between pea cultivars and the soaking treatments for the number of bacterial nodules/plant and nitrogenase activity. The obtained data indicated that R/T treatments with the tested cultivars had the highest values. In concern of silicate x cultivars, data showed noticeable increments with Sugar gum x MgSiO₃ (4g/L) for number of bacterial nodules/plants for both seasons with averages of 11.333 and 10.333, respectively and 0.591 for nitrogenase activity in the first season only.

Table 6. Effect of pea cultivars, seed soaking treatments and their interaction on number of root bacterial nodules/plant, nitrogenase activity and seed protein content g kg⁻¹ seed during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

Parameter	Number of bacterial nodules / plants				Nitrogenase activity				Seed protein content g /kg seed			
	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean
Treatment	1st season											
Mg SiO ₃ 2g/L	6.67	4.67	6.67	6.00 B	0.40	0.37	0.56	0.44 B	107.50	105.50	109.38	107.46 B
Mg SiO ₃ 4g/L	9.33	7.33	11.33	9.33 A	0.49	0.59	0.59	0.56 A	111.88	108.58	116.33	112.27 A
K ₂ SiO ₃ 2g/L	4.00	3.33	4.67	4.00 C	0.32	0.31	0.33	0.32 D	105.63	103.13	109.38	106.05 B
K ₂ SiO ₃ 4g/L	6.67	5.33	7.67	6.56 B	0.38	0.34	0.39	0.37 C	109.38	105.00	111.88	108.75 B
R/T 3g/L	10.00	8.00	14.33	10.78 A	0.55	0.39	0.57	0.51 A	111.88	109.38	116.25	112.50 A
Control	3.33	2.67	3.67	4.00 C	0.27	0.25	0.29	0.27 E	101.25	101.88	102.50	101.88 C
C. mean	6.67 AB	5.22 B	8.06 A		0.40	0.38	0.46		107.92	105.58	110.95	
LSD 5% C	1.42				0.13 (NS)				4.46 (NS)			
T	1.11				0.07				4.25			
C x T	2.32				0.08				4.94			
	2nd season											
Mg SiO ₃ 2g/L	4.33	3.67	5.67	4.56 D	0.40	0.49	0.39	0.42 AB	108.13	104.38	108.75	107.09 C
Mg SiO ₃ 4g/L	7.67	6.33	10.33	8.11 B	0.39	0.56	0.24	0.40 ABC	111.88	108.13	115.63	111.88 A
K ₂ SiO ₃ 2g/L	3.00	2.33	3.67	3.00 E	0.31	0.32	0.32	0.32 BC	104.00	103.75	113.75	107.17 BC
K ₂ SiO ₃ 4g/L	5.67	4.33	6.67	5.56 C	0.38	0.37	0.34	0.37 ABC	108.75	105.63	114.38	109.59 AB
R/T 3g/L	9.00	6.67	13.33	9.67 A	0.36	0.55	0.55	0.49 A	110.00	110.63	115.63	112.09 A
Control	2.33	1.67	2.67	2.22 F	0.28	0.27	0.27	0.27 C	101.25	101.88	102.50	101.88 D
C. mean	5.33 B	4.17 C	7.06 A		0.35	0.43	0.35		107.34 AB	105.73 B	111.77 A	
LSD 5% C	0.60				0.11 (NS)				4.99			
T	0.66				0.11				3.04			
C x T	0.77				0.13				3.54			

2.2. d. Effect of seed soaking treatments on cultivars and their interaction for chlorophyll content:

Data presented in Table (7) clear that the cultivar Sugar gum had the highest value followed by Master B and Blmewar for both seasons with averages of 0.339, 0.294 and 0.281(mg/g leaves fresh weight) for the first season and 0.298, 0.287 and 0.281 (mg/g leaves fresh weight) for the second season, respectively. In case of soaking treatments R/T (3g/L) showed the highest chlorophyll a value followed by MgSiO₃ (4g/L) and K₂SiO₃ (4g/L) for both seasons with averages of 0.479, 0.403 and 0.337 (mg/g leaves fresh weight) for the first seasons and 0.455, 0.355 and 0.355(mg/g leaves fresh weight) for the second once, respectively. In case of chlorophyll b content, the same arrangement cleared with averages of 0.123, 0.120 and 0.116 (mg/g leaves fresh weight) for the first season and 0.127, 0.118 and 0.111(mg/g leaves fresh weight) for the second season, respectively. The results of chlorophyll a and b reflected on the content of the total chlorophyll (chlorophyll a+b). For the interaction effect, R/T (3g/L) x the three tested pea cultivars had the highest value of chlorophyll a, b and a + b. At the same time, Sugar gum x MgSiO₃ (4g/L) had considerable increase for chlorophyll a, b and a + b for both seasons.

Table7. Effect of pea cultivars, seed soaking treatments and their interaction on chlorophyll a, b and a+b content (mg/g leaves fresh weight) during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

Parameter	Chlorophyll A				Chlorophyll B				Chlorophyll A+B			
	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean
Treatment	1 st season											
Mg SiO ₃ 2g/L	0.23	0.22	0.33	0.26 D	0.11	0.10	0.11	0.11 B	0.33	0.32	0.43	0.36 B
Mg SiO ₃ 4g/L	0.39	0.35	0.47	0.40 B	0.12	0.11	0.13	0.12 A	0.51	0.46	0.60	0.52 A
K ₂ SiO ₃ 2g/L	0.24	0.22	0.22	0.23 E	0.10	0.10	0.10	0.10 D	0.35	0.32	0.33	0.33 B
K ₂ SiO ₃ 4g/L	0.34	0.33	0.34	0.34 C	0.12	0.10	0.13	0.12 A	0.46	0.43	0.46	0.45 A
R/T 3g/L	0.45	0.44	0.55	0.48 A	0.13	0.13	0.11	0.12 A	0.58	0.57	0.66	0.60 A
Control	0.13	0.12	0.13	0.13 F	0.09	0.09	0.10	0.09 C	0.22	0.21	0.23	0.22 C
C. mean	0.29 B	0.28 B	0.34 A		0.11 A	0.10 B	0.11 A		0.41	0.38	0.45	
LSD 5% C	0.03				0.01				0.11 (NS)			
T	0.02				0.005				0.09			
C x T	0.02				0.01				0.11			
	2 nd season											
Mg SiO ₃ 2g/L	0.23	0.22	0.23	0.22 C	0.10	0.10	0.11	0.10 D	0.33	0.32	0.33	0.33 C
Mg SiO ₃ 4g/L	0.33	0.35	0.38	0.36 B	0.11	0.12	0.12	0.12 B	0.44	0.47	0.50	0.47 B
K ₂ SiO ₃ 2g/L	0.22	0.22	0.22	0.22 C	0.11	0.10	0.10	0.10 D	0.33	0.32	0.32	0.32 C
K ₂ SiO ₃ 4g/L	0.34	0.34	0.39	0.36 B	0.12	0.10	0.11	0.11 C	0.46	0.44	0.50	0.47 B
R/T 3g/L	0.49	0.44	0.45	0.46 A	0.13	0.13	0.13	0.13 A	0.62	0.57	0.58	0.59 A
Control	0.12	0.12	0.13	0.12 D	0.09	0.10	0.09	0.09 E	0.22	0.22	0.21	0.21 D
C. mean	0.29 B	0.28 C	0.30 A		0.11	0.11	0.11		0.40 B	0.39 C	0.41 A	
LSD 5% C	0.01				0.006 (NS)				0.007			
T	0.02				0.007				0.02			
C x T	0.02				0.01				0.02			

2.5. Effect of seed pea soaking treatments on cultivars and their interaction for root lignin content:

Data in Table (8) show that Sugar gum roots had the highest root lignin content followed by Master B and Blmewar with averages of 1.125, 0.999 and 0.884% in the first season and 0.942, 0.934 and 0.833% in the second season, respectively. For pea seed soaking, data in Figure (2) appear that R/T treatment had the highest lignin content followed by MgSiO₃ (4g/L), MgSiO₃ (2g/L) and K₂SiO₃ (4g/L) for both seasons with averages of 1.360, 0.988, 0.951 and 0.907% for the first season and 1.127, 0.967, 0.927 and 0.888% for the second season, respectively.

Table8. Effect of seed pea soaking treatments, cultivars and their interaction on root lignin content during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

Root lignin content				
Cultivar	Master B	Blmewar	Sugar gum	Treatment mean
Treatment	1 st season			
Mg SiO ₃ 2g/L	0.99	0.85	1.01	0.95 BC
Mg SiO ₃ 4g/L	1.04	0.89	1.04	0.99 B
K ₂ SiO ₃ 2g/L	0.84	0.83	0.88	0.85 C
K ₂ SiO ₃ 4g/L	0.87	0.87	0.99	0.91 BC
T/R 3g/L	1.30	1.01	1.77	1.36 A
Control	0.95	0.85	1.07	0.96 BC
Cultivar mean	1.00	0.88	1.13	
LSD 5% C	0.22 (NS)			
T	0.12			
C x T	0.15			
	2 nd season			
Mg SiO ₃ 2g/L	0.98	0.84	0.96	0.93 BC
Mg SiO ₃ 4g/L	1.01	0.86	1.03	0.97 B
K ₂ SiO ₃ 2g/L	0.83	0.82	0.86	0.84 D
K ₂ SiO ₃ 4g/L	0.85	0.84	0.97	0.89 CD
T/R 3g/L	1.27	1.00	1.11	1.13 A
Control	0.66	0.63	0.72	0.67 E
Cultivar mean	0.93	0.83	0.94	
LSD 5% C	0.16 (NS)			
T	0.08			
C x T	0.09			

In case of interaction between pea cultivars and seed soaking, it can be noticed that the treatment of R/T (3g/L) x the three tested cultivars had the highest lignin %. In this concern, in the first season Master B x MgSiO₃ (4g/L) had a considerable increase followed by Sugar gum x MgSiO₃ (4g/L) with averages of 1.040 and 1.037%, respectively and in the second season Sugar gum x MgSiO₃ (4g/L) followed by Master B x MgSiO₃ (4g/L) with averages of 1.027 and 1.010%, respectively.

2.6. Effect of seed pea soaking treatments on cultivars and their interaction for the activities of polygalacturonase (PG) and Cx-cellulase (Cx):

From the data presented in Table (9) it can be noticed that Sugar gum had the least values for both PG and Cx-cellulase activities with averages of 37.628 and 37.53 (% loss in viscosity) followed by Blmewar and Master B, respectively. In case of pea soaking treatments, in spite of R/T (3g/L) treatment sharply decreased the production of the both enzymes; PG and Cx with averages of 22.22 and 24.50 (% loss in viscosity), respectively, magnesium and potassium silicate had significant effects in reducing the production of the previous two enzymes compared to control treatments. MgSiO₃ (4g/L) produced PG (40.17%) and Cx (42.87%) viscosity loss followed by MgSiO₃ (2g/L) with 44.88% and 47.13% compared to untreated control which produced 84.48 and 85.50%, respectively.

In this respect, the interaction between cultivars and soaking treatments had a significant decrease in PG and Cellulase activities. Although R/T (3g/L) treatment caused considerable decrease production of PG and Cx (% loss in viscosity) with the three tested cultivars, MgSiO₃ and K₂SiO₃ decreased the production of the previous enzymes. Sugar gum x MgSiO₃ (4g/L) had 25.12% for PG followed by Sugar gum x MgSiO₃ (2g/L), Sugar gum x K₂SiO₃ (4g/L) and Sugar gum x K₂SiO₃(2g/L) with averages of 26.19, 37.25 and 39.33%, respectively. On the other hand, these results were close to Cx-Cellulase with the same arrangement with averages of 29.14, 30.22, 30.42 and 35.47%, respectively.

Table9. Effect of pea cultivars, seed soaking treatments and their interaction on polygalacturonase (PG) and Cx – cellulose activity (loss of viscosity (%)) after 60 min) of some pea varieties during two successive seasons; 2021/2022 and 2022/2023 under field conditions.

Parameter	polygalacturonase activity				Cx – cellulose activity			
	Master B	Blmewar	Sugar gum	T.mean	Master B	Blmewar	Sugar gum	T.mean
Treatment								
Mg SiO ₃ 2g/L	60.22	48.22	26.19	44.88 D	65.25	45.91	30.22	47.13 C
Mg SiO ₃ 4g/L	55.27	40.11	25.12	40.17 D	59.30	40.18	29.14	42.87 C
K ₂ SiO ₃ 2g/L	73.62	51.14	39.33	54.70 B	83.15	52.13	35.47	56.92 B
K ₂ SiO ₃ 4g/L	62.61	49.22	37.25	49.69 C	80.13	49.99	30.42	53.51 B
R/T 3g/L	25.17	23.44	17.33	21.98 E	29.53	23.71	20.25	24.50 D
Control	90.13	82.77	80.55	84.48 A	85.61	91.22	79.66	85.50 A
C. mean	61.17 A	49.15 B	37.63 C		67.16 A	50.52 B	37.53 C	
LSD 5% C		9.68			7.92			
T		4.79				7.84		
C x T		5.58				9.12		

2.7. Relationship between pea seed soaking treatments, cultivars and their interaction and pre- and post - emergence damping off:

In the case of the three tested pea cultivars, data illustrated in Figure (1) show that Master B had higher values of polygalacturonase and cellulase enzymes (PG, Cx) than Blmewar and Master B. Also, data clear that (PG) and Cx enzymes were in parallel with the incidence of damping – off disease (average of two seasons) where the activity of any enzyme from them decreases with the decreasing of damping – off. R/T (3g/L) treatment had the least PG, Cx, pre- and post - emergence damping – off with averages of 22.22, 24.50, 7.75 and 4.40%, respectively. At the same time MgSiO₃ and K₂SiO₃ had significant decrease for PG and Cx-cellulase production compared to control treatment. MgSiO₃ (4g/L) showed 40.17 and 42.87 followed by MgSiO₃(2g/L) with 44.88 and 47.13 loss in viscosity%, respectively. For the interaction effect, the results are in agreement with that of soaking treatments and the cultivars where the best values were related to the cultivar Sugar gum x MgSiO₃ (4 and 2g/L) and K₂SiO₃ (4 and 2g/L), respectively.

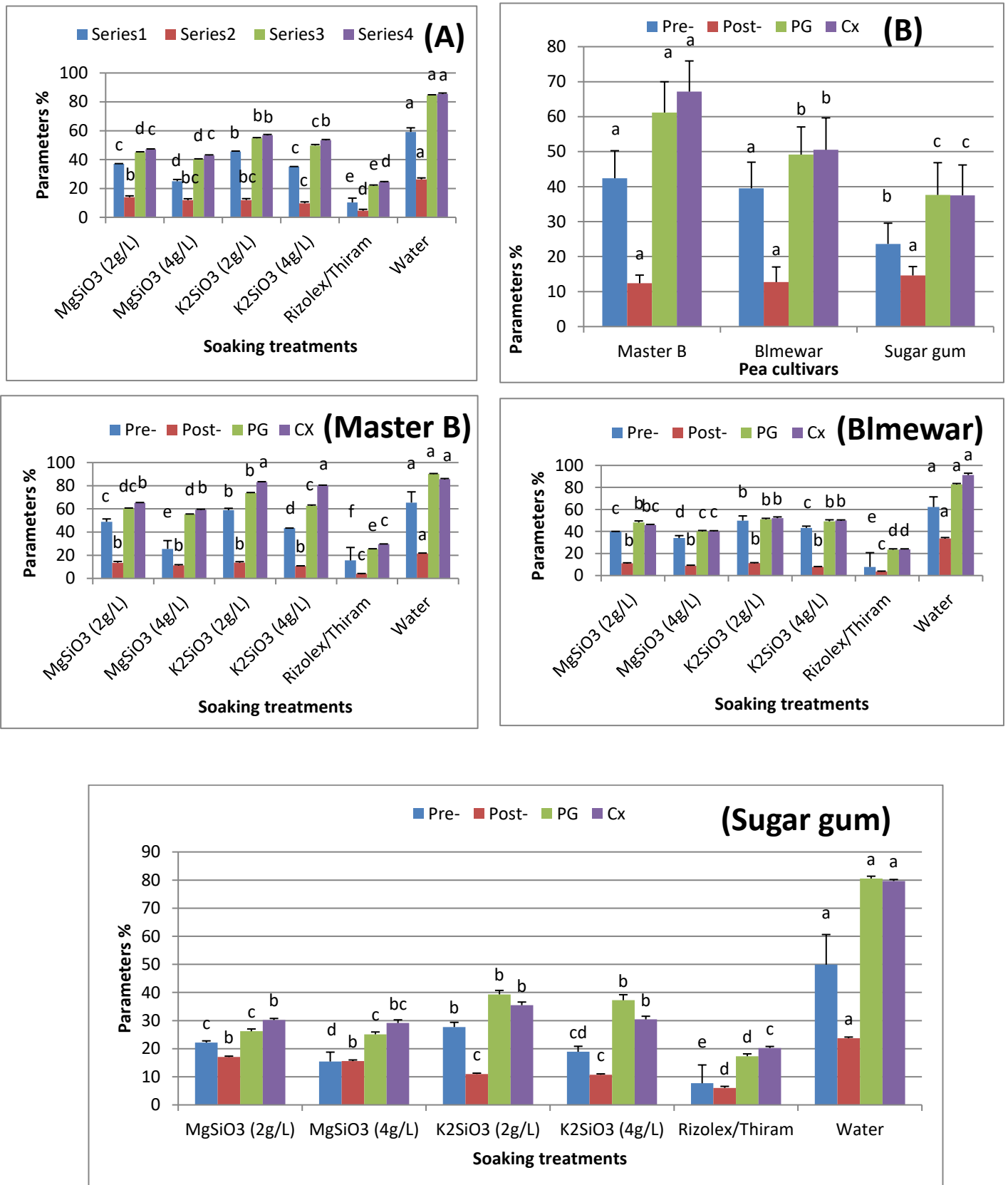


Fig.1. Effect of soaking pea seeds on different sources of silicate (A) on pea cultivars (B) and their interactions for pea pre-, post-emergence damping-off , polygalacturonase and cellulose activities(loss in viscosity%).

DISCUSSION

The investigation identified *Fusarium oxysporum*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, and *Pythium ultimum* as the fungi responsible for root rot in pea plants, utilizing the methodologies established by Barnett and Hunter (1972) and Van der Plaats-Niterink (1981). These findings are consistent with those of (Grünwald *et al.* (2004), who also determined that soil-borne fungi cause damping-off and root rot. Pathogenicity tests confirmed that these fungi were pathogenic to the pea cultivars Master B, Blmewar, and Sugar Gum, with varying levels of disease severity observed among these cultivars (Muhanna *et al.*, 2018). The degree of disease reduction varied among the cultivars, with Sugar Gum exhibiting the least incidence of damping-off for both seasons, followed by Blmewar and Master B. This indicates that the effectiveness of silicon treatments can be attributed to the inherent genetic resistance factors present in each cultivar, which influence their ability to withstand fungal attacks (Ney *et al.*, 2013).

Damping-off disease affects seedling emergence and survival, leading to substantial yield losses. The use of silicon (Si) as a seed treatment has gained attention due to its potential to enhance plant resistance against various stresses, including pathogen attacks (Giovanna *et al.*, 2024). Although silicon is not considered an essential nutrient for plant growth, its role in enhancing plant resistance to diseases is significant (Debona *et al.*, 2017) and that is through soaking seeds in a silicon solution before planting aims to enhance seedling vigor and improve resistance to soil-borne pathogens (Debona *et al.*, 2017). Additionally, silicon can induce systemic resistance in plants, activating various defense mechanisms (Putra *et al.*, 2020).

The significant reduction in the incidence of damping-off disease in silicon-treated seeds underscores the potential of silicon-based treatments as eco-friendly and sustainable alternatives to conventional fungicides. Although Rizolex/Thiram, commercial fungicides, remain highly effective against damping-off disease—caused by various soil-borne pathogens including fungi and oomycetes (e.g., *Pythium*, *Rhizoctonia*, and *Fusarium* spp.)—the use of magnesium silicate (MgSiO₃) and potassium silicate (K₂SiO₃) offers a promising approach to disease management. This approach potentially reduces reliance on chemical fungicides and mitigates their environmental impact. Several researchers decided that the efficacy of these silicon treatments can be attributed to several mechanisms (Bardisi, 2004; Vivancos *et al.*, 2015; Abd El-Gawad *et al.*, 2017; Putra *et al.*, 2020).

Silicon is absorbed by plants as monosilicic acid (H₄SiO₄) and is deposited in tissues, where it helps to fortify cell walls and form physical barriers against pathogen invasion. Additionally, silicon can regulate the expression of genes linked to plant defense, resulting in the production of phenolic compounds, lignin, and other molecules involved in defense. Moreover, silicon enhances photosynthetic efficiency, improves nutrient uptake, and bolsters overall plant health, making plants more resilient to pathogen attacks. Soaking pea seeds in two sources of silicate salts significantly increased root length, root fresh weight, root dry weight, seed yield per plant, number of root bacterial nodules per plant, nitrogenase activity, and protein content compared to untreated control, due to silicon treatments which not only reduced disease incidence but also promoted better seedling vigor and growth and the treated seedlings were more robust and had higher survival rates, contributing to overall improved plant health (Fortunato *et al.*, 2012; Van *et al.*, 2013; and Shalaby *et al.*, 2017). The protective effects of silicon are attributed to both physical and biochemical mechanisms. Since, silicon strengthens cell walls, creating a physical barrier against pathogen entry and, silicon induces the production of defense-related compounds such as chlorophyll content, root lignin content, and pathogenesis-related proteins activities of (polygalacturonase (PG) and Cx-cellulase (Cx)), enhancing the plant's immune response (Liang, 1999; Chen *et al.*, 2014; Rodrigues *et al.*, 2015; Vivancos *et al.*, 2015).

Legumes (Fabaceae) form a symbiotic relationship with nitrogen-fixing bacteria (rhizobia) in their root nodules, enabling them to fix atmospheric nitrogen. Some researchers suggested that silicon promotes this symbiosis because silicon enhances the legume-rhizobia interaction, increasing root nodulation, bacteroid numbers, and nitrogen fixation across various legume species. Given the global importance of legumes, silicon could significantly enhance legume health and productivity, offering substantial environmental benefits (Vessey, 1994; Xia, *et al.*, 2017; Steiner *et al.*, 2018; Putra *et al.*, 2020). Various research groups referred that understanding the interactions between seed soaking treatments and genetic resistance is essential for optimizing disease control in pea cultivation (Biancardi, 2005; Bongard, 2016; Brantner and Chanda, 2018; Eliana *et al.*, 2023). Damping-off, caused by soil-borne pathogens, significantly impacts seedling emergence and early growth. Effective management involves using seed treatments and selecting resistant cultivars. Seed treatments, particularly fungicides, provide a protective barrier, while nutrient solutions like silicon strengthen seedlings. The effectiveness of these treatments varies with the cultivar's genetic makeup, with Sugar Gum showing the highest resistance and benefiting most from treatments, while Blmewar and Master B exhibit moderate resistance.

CONCLUSION

This study assessed three pea cultivars' responses to damping-off disease caused by various fungal pathogens. Field trials showed that Sugar Gum had the lowest damping-off rates and best growth, while Blmewar had the highest seed weight per plant. Rizolex/Thiram was the most effective treatment, but magnesium silicate ($MgSiO_3$) at 4 g/L also significantly reduced damping-off and improved growth. Selecting resistant cultivars and using effective treatments as silicon that can manage damping-off and enhance pea yield and quality.

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