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# Study of endolichenic *Trichoderma* resistance to abiotic stresses and their potential for growth promotion of tomato plants (*Solanum lycopersicum*)

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#### ABSTRACT

This study focused on the isolation of *Trichoderma* strains from lichens collected from Acacia trees located in Misserghine, Algeria, the evaluation of their tolerance to abiotic stresses at different pH (4.5, 6 and 8) and temperature (15 °C, 20 °C, 25 °C and 35 °C) and their potential use to promote the germination and growth of tomato plants (*Solanum lycopersicum*). Five *Trichoderma* strains were isolated and morphologically identified. The results showed that the optimal growth of strains was obtained at pH 4.5 and 25 °C with variability between strains, notably S5 which showed better tolerance to alkaline pH. The effect of these strains on the germination of tomato plants showed that the majority of the strains significantly improved the germination rate (80 to 86% versus 70% for the control) and the seed vigor index. Strains S2 and S4 recorded the best results, almost doubling the vigor index compared to the control. The evaluation of vegetative growth of tomato plants after four weeks confirmed these positive results, especially for strain S4 which significantly increased the length of roots (7.84 cm) and stems (16.38 cm) compared to the control (3 cm and 13.98 cm, respectively). However, strain S3 showed negative effects on plant development by reducing the seed vigor index to 314.88 compared to the control with S.V.I of 756, decreasing the stems length to 9.42 cm and exhibiting no significant effect on root growth.

Keywords: abiotic stress, acacia tree, growth promotion, lichens, Trichoderma, tomato (Solanum lycopersicum).

### INTRODUCTION

The agricultural sector in Algeria is crucial for the country's economic development and food security. Since the launch of the National Plan for Agricultural and Rural Development (PNDAR) in 2000, agricultural production has continued to increase, contributing to around 12% of the national GDP. With a total agricultural area of 42.4 million hectares, of which 8.458 million hectares are usable, the agricultural sector provides livelihoods to about 21% of the national population and occupies an important place in the economy, constituting 5 to 7% of the total value of the country exports in 2023 (Mellab, 2025). Improving crop production relies mainly on the use of chemicals, such as fertilizers and pesticides, which improve agricultural yield and prevent damage from plant pathogenic microorganisms. However, the massive use of chemicals is not without consequences; their long-term use can cause significant pollution of soil, water and air, and pose risks to human health (Kredics et al., 2024; Menadi et al., 2024).

Faced with these challenges, it has become essential to promote more sustainable agricultural practices based on ecological alternatives, such as the use of biological treatments which increase agricultural yield while guaranteeing soil quality.

Trichoderma spp. are widely recognized as non-pathogenic plant symbionts capable of colonizing plant roots and establishing a beneficial interaction with their hosts. This interaction results in an improvement in plant biomass and effective protection against several phytopathogens. As a result, *Trichoderma* exerts a direct bioprotective effect through the mechanisms of mycoparasitism, antibiosis, competition for space and nutrients and the production of lytic enzymes and indirectly by stimulating the natural defense system of plants and inducing systemic resistance. This diversity of biocontrol mechanisms has imposed *Trichoderma* as a promising and sustainable alternative to chemical phytosanitary products (Ferreira and Musumeci, 2021; Tyskiewicz et al., 2022; Guzmán-Guzmán et al., 2023)

Although *Trichoderma* species are widely used as beneficial agricultural bioinoculants, some studies have revealed that their interactions with plants are more complex and *Trichoderma*plant interactions are highly specific with consequences ranging from beneficial to neutral or even harmful effects on plant development (Tucci et al., 201; Pozo et al., 2024; Pfordt et al., 2025).

The success of these interactions depends on multiple factors, including the host plant genotype, the fungal strain specificity and environmental conditions (Santos et al., 2020). Some strains may reduce root development or decrease overall plant growth highlighting the critical importance of comprehensive strain characterization and compatibility assessment before their implementation in agricultural applications.

In this context, the presented study aimed to isolate and characterize *Trichoderma* strains from the lichens collected in Misserghine region in Algeria and to evaluate their potential as plant growth promoters. Their tolerance to abiotic stresses (pH and temperature) was investigated and their effect on tomato seed germination as well as plant development with the goal of identifying promising strains for agronomic applications were assessed.

## MATERIALS AND METHODS

### Sample collection

Lichen samples were collected from Acacia trees (Figure 1) located in Misserghine, Northwest of Algeria (Figure 2). The collection was performed using sterile tweezers and a knife, carefully scraping the bark of the trees. The collected samples were then placed in paper bags and immediately transported to the laboratory.

### Isolation of Trichoderma

To isolate *Trichoderma*, the collected samples were rinsed with sterile distilled water, then immersed in 95% ethanol for 30 seconds, in 0.5%



Figure 1. Lichens on Acacia tree

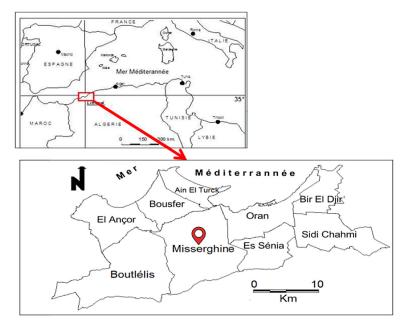


Figure 2. Location of Misserghine - coordinates 35°37'N, 0°44'W

sodium hypochlorite for 2 seconds, and in 70% ethanol for 30 seconds followed by three rinses with sterile distilled water (Oh et *al.*, 2020). The disinfected lichen fragments were placed on PDA medium supplemented with 5 mg/L gentamicin. The Petri dishes were then incubated at 25 °C.

### Morphological identification of *Trichoderma isolates*

The morphological identification of the isolates was based on macroscopic observation of cultural characteristics and microscopic examination after 7 days of incubation on PDA medium. The identification of isolates at the genus level was conducted using the identification key of Barnett and Hunter (1998).

# Screening for abiotic stress tolerance of isolates

The abiotic stress tolerance of *Trichoderma* isolates was evaluated based on their ability to grow under varying pH and temperature conditions. To assess the effect of pH, 8 mm inoculum discs, taken from one week old cultures, were inoculated at the center of Petri dishes containing PDA medium adjusted to three pH levels 4.5, 6 and 8. The plates were incubated at 25 °C for 8 days. Temperature tolerance of isolates was tested by cultivating the isolates on PDA medium at 15 °C, 20 °C, 25 °C and 30 °C. Each experimental

condition was performed in triplicate for each strain. The radial growth of the colonies was measured every 48 hours.

# Effect of *Trichoderma* strains on tomato seed germination

#### Disinfection and seed preparation

Tomato seeds (*Solanum lycopersicum*, variety Marmande) were disinfected by soaking in a 2% sodium hypochlorite solution, followed by rinsing with sterile distilled water. The seeds were then dried on a sterile filter paper before use.

#### Preparation of Trichoderma spore suspension and seed treatment

*Trichoderma* strains were cultured on PDA medium at 25 °C for 15 days. Spores were harvested by scraping the surface of the mycelial cultures. The resulting mixture was filtered through a Whatman No. 1 filter paper, and the spore suspension density was adjusted to  $2 \times 10^8$  spores/ml using Malassez cell. The disinfected seeds were soaked in the *Trichoderma* spore suspension and air dried overnight under a laminar flow hood. The seeds treated with sterile distilled water served as control (Rubio et *al.*, 2012).

#### Germination test

The treated and control seeds were placed in Petri dishes containing two layers of sterile filter paper moistened with sterile distilled water. Each dish contained 25 seeds, with four replicates for each treatment. The dishes were incubated in darkness at 25 °C and the number of germinated seeds was counted every 24 hours. The germination percentage was calculated at the end of the experiment once no further germination was observed after two successive counts. The root and hypocotyl lengths were measured on the 21<sup>st</sup> day of incubation to calculate the seed vigor index (S.V.I.). S.V.I. was determined using the formula of Abdul Baki and Anderson (1973) : S.V.I. = germination percentage x (root length + hypocotyl length)

# Effect of *Trichoderma* strains on tomato plant growth

An *in-vivo* assay was used to evaluate the ability of *Trichoderma* strains to promote the tomato plant growth. Tomato seeds, treated and untreated with *Trichoderma*, were sown in pots at a rate of 3 seeds per pots, with 5 pots per treatment. Each pot was filled with a sterile substrate composed of soil and peat (2:1, v/v). The plants were maintained at 22 °C and watered every two days with distilled water. After four weeks, the stem and root lengths of each plant were measured to assess the effect of *Trichoderma* on growth (Rubio et *al.*, 2012).

## **Statistical analysis**

Statistical analyses were performed using Python 3 and Jupyter software. The Shapiro-Wilk test and Levene test were used to verify the normality and homogeneity of variances. The ANO-VA (Analysis of Variance) and Tukey post-hoc test were used to compare the growth of fungal isolates at different pH and temperature values. P-values were included to strengthen the results, with a significance threshold of 5% (p < 0.05) to ensure the reliability of the conclusions. The objective was to assess whether different pH levels and different temperatures significantly influence the growth of fungal isolates. For this purpose, the growth measurements obtained under various pH conditions and temperature settings were compared. Specifically, an ANOVA test was performed to determine if there were statistically significant differences between the groups (different pH levels and different temperatures). When the ANOVA indicated a significant effect, we conducted a post-hoc Tukey test to precisely identify

which pairs of groups differed from each other. In the case of pH, the groups compared were: pH 4.5 vs pH 6, pH 4.5 vs pH 8, and pH 6 vs pH 8. In the case of temperature, the groups compared were: 15 °C vs 20 °C, 15 °C vs 25 °C, 15 °C vs 35 °C, 20 °C vs 25 °C, 20 °C vs 35 °C, and 25 °C vs 35 °C. This methodological approach enables to rigorously evaluate the impact of pH and temperature on fungal growth.

## RESULTS

#### Isolation and identification of Trichoderma

Five *Trichoderma* isolates were obtained from the lichen samples. Their rapid growth and the greenish appearance of their colonies allowed their assignment to the genus *Trichoderma* (Figure 3). This identification was confirmed through microscopic observation of characteristic morphological structures including hyaline non verticillate and branched conidiophores, ovoid phialides producing unicellular, hyaline, round, smooth and green conidia which were formed in masses at the tips of the phialides.

# Screening for abiotic stress tolerance of isolates

## Effect of pH

The result showed that *Trichoderma* strains exhibited optimal growth at an acidic pH of 4.5. Growth gradually decreased as the pH increased, reaching a minimum at pH 8. Strain S5 demonstrated the best growth at pH 6 (3.8 cm) and maintained relatively high growth at pH 8 (2.1 cm), indicating greater tolerance to pH variation. In contrast, the growth of the other strains was reduced at pH 8 with colony diameters ranging from 1.03 cm to 1.4 cm (Figure 4).

According to the statistical study, Table 1 presents the descriptive statistics of the growth measurements of fungal isolates at different pH values, including the minimum and maximum values, the mean, the median, the mode, and the standard deviation.

Before applying a parametric test, it is essential to verify whether the data follow a normal distribution. In the considered case, the Shapiro-Wilk test was used, which is particularly suitable for small

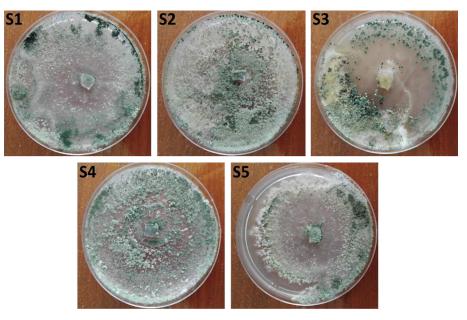


Figure 3. Trichoderma isolates from the lichen samples

sample sizes. Once normality was confirmed, the homogeneity of variances was assessed using Levene's test, which yielded a p-value of 0.997, indicating that the variances are homogeneous.

Then, a one-way ANOVA was performed to compare the growth of fungal isolates across different pH levels. The analysis revealed a p-value of 0.004, suggesting a significant effect of pH on growth. To precisely identify the differences between pH levels, a Tukey post-hoc test was conducted, the results of which are presented in Table 2.

The results show that there is no significant difference between pH 4.5 and pH 6, indicating similar growth of the fungal isolates under these two conditions. In contrast, significant differences were observed between pH 4.5 and pH 8, as well as between pH 6 and pH 8, with markedly lower growth at pH 8. Hence, pH 8 has a significantly

negative effect on the growth of the fungal isolates, while no notable difference was observed between pH 4.5 and pH 6 (Figure 5).

### Effect of temperature

The mycelial growth of *Trichoderma* strains was also affected by temperature. The optimal temperature was 25 °C, at which all strains achieved maximum growth. A doderate decrease in growth was observed at 20 °C, while a significant reduction in growth was noted at 15 °C. No growth was detected at 35 °C for any of the strains, indicating intolerance to high temperature (Figure 6).

According to the statistical study, Table 3 presents the descriptive statistics of the growth measurements of the fungal isolates under the different temperatures tested. It includes the

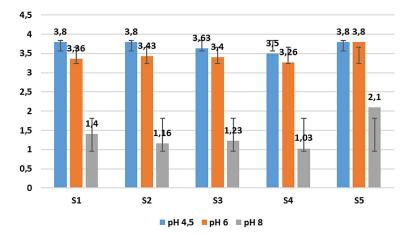


Figure 4. Macroscopic aspect of Trichoderma strains

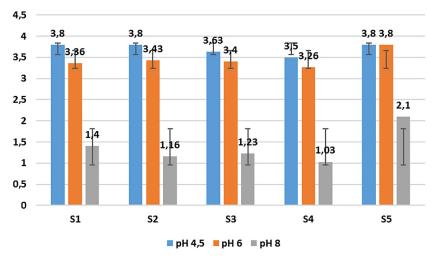


Figure 5. Effect of pH on the radial growth of Trichoderma strains

Table 1. Descriptive statistics of the growth of Trichoderma strains at different pH values

Parameter	pH 4.5	рН 6	pH 8
Min	3.5	3.26	1.03
Mean	3.706	3.45	1.384
Median	3.8	3.4	1.23
Mode	3.8	/	/
Standard deviation	0.1366748	0.2059126	0.42193601
Max	3.8	3.8	2.1

 Table 2. Tukey post-hoc results

Comparaison	Adjusted P-value	Conclusion
pH 4.5 vs pH 6	0.3555	No significant difference
pH 4.5 vs pH 8	0.0000	Significant difference
pH 6 vs pH 8	0.0000	Significant difference

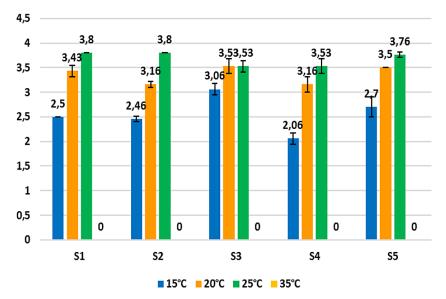


Figure 6. Effect of temperature on the radial growth of *Trichoderma* strains

minimum and maximum values, mean, median, mode, and standard deviation for each temperature condition.

To determine the appropriate type of test to apply (parametric or non-parametric), it was first necessary to verify whether the data followed a normal distribution. For this purpose, the Shapiro-Wilk test was performed using Python programming language, which is particularly suitable for small sample sizes.

After confirming normality (p-value = 0.084), we assessed the homogeneity of variances using Levene's test, which yielded a p-value of 0.882, indicating that the variances are homogeneous.

Then, an ANOVA test was performed to analyze the effect of temperature on the growth of the fungal isolates. The analysis revealed a p-value of 0.000, suggesting a significant effect of temperature.

To precisely identify the differences between the tested temperatures, a Tukey post-hoc test was conducted, the results of which are presented in Table 4.

The results show that there is no significant difference between 20 °C and 25 °C, indicating similar growth of the fungal isolates at these two temperatures. In contrast, significant differences were observed between 15 °C and all other temperatures, as well as between 35 °C and the lower temperatures, suggesting a marked impact of temperature on growth. These findings indicate that temperature significantly influences the growth of fungal isolates, with notable differences observed between several temperature levels, except between 20 °C and 25 °C, where growth remains similar.

# Effect of *Trichoderma* strains on tomato seed germination

The germination rate of tomato seeds (Table 5) showed a significant improvement in those treated with different *Trichoderma* strains compared to the untreated control seeds (T). The control seeds recorded a germination rate of 70%, while the treated seeds showed higher rates, ranging from 80% to 86%. Strain S4 exhibited the highest germination rate at 86%, followed by S2 and S5 at 84%, then S3 at 82% and S1 at 80%.

The evaluation of the seed vigor index (S.V.I.) (Table 5, Figures 7 and 8) revealed significant differences between the *Trichoderma* treated seeds and the untreated control. The control recorded an S.V.I. of 756, while treated seeds generally showed higher values except for strain S3. Strains S4 and S2 demonstrated the most positive effects with S.V.I. values of 1 553.16 and 1 533.84 respectively, nearly double the control value. These results are particularly attributed to a significant increase in root length, reaching 11.56 cm for S4 and 11.72 cm for S2, compared to 5.64 cm for the control. Strains S1 and S5 also showed positive

Parameter	15 °C	20 °C	25 °C	35 °C
Min	2.06	3.16	3.53	0
Mean	2.556	3.356	3.684	0
Median	2.5	3.43	3.76	0
Mode	1	3.16	3.8	0
Standard deviation	0.36507533	0.18256506	0.14152738	0
Max	3.06	3.53	3.8	0

Table 3. Descriptive statistics of the growth of Trichoderma strains at different temperatures

Table 4. Tukey post-hoc results

Comparaison	Adjusted P-value	Conclusion
15 vs 20	0.0001	Significant difference
15 vs 25	0.0000	Significant difference
15 vs 35	0.0000	Significant difference
20 vs 25	0.1170	No significant difference
20 vs 35	0.0000	Significant difference
25 vs 35	0.0000	Significant difference

Strain	Germination rate	Hypocotyl and root length (cm)		S.V.I.
		Hypocotyl	Root	5.v.i.
Т	70%	5.16	5.64	756
S1	80%	6.14	9.18	1 225.6
S2	84%	6.54	11.72	1 533.84
S3	82%	1.46	2.38	314.88
S4	86%	6.5	11.56	1 553.16
S5	84%	6.36	9.7	1 349.04

Table 5. Effect of Trichoderma strains on tomato seed germination

effects with S.V.I. values of 1 225.6 and 1 349.04 respectively. However, strain S3 showed lower value even less than the control with an S.V.I. of 314.88 mainly due to reduced growth of both hypocotyl (1.46 cm) and root (2.38 cm).

# Effect of *Trichoderma* strains on tomato plant growth

The effect of *Trichoderma* strains on tomato plant growth was assessed after four weeks of cultivation. The results showed that most *Trichoderma* treatments enhanced both root and stem growth compared to the control. Strain S4 demonstrated the most significant growth promoting effect, with root length reaching 7.84 cm and stem length of 16.38 cm compared to 3.4 cm and 13.98 cm, respectively, for the control plants. Strain S2 also showed notable growth enhancement with root length of 5.74 cm and stem length of 15.8 cm, followed by strain S5 (root: 5 cm, stem: 15.04 cm) and S1 (root: 4.84 cm, stem:

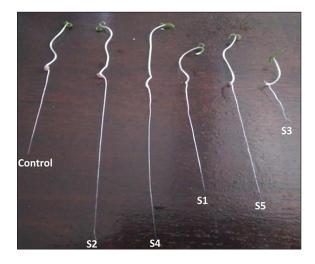


Figure 7. Effect of *Trichoderma* strains on root and hypocotyl length

15.42 cm). However, strain S3 exhibited no significant effect on root growth and had a negative impact on stem growth, with lengths of 3.22 cm and 9.42 cm respectively (Figures 9 and 10).

### DISCUSSION

Five strains of *Trichoderma* were isolated from the lichen samples. The influence of pH and temperature on the mycelial growth of these strains was investigated in the conducted study. The optimal growth of *Trichoderma* strains was observed between pH 4.5 and 6, reflecting its natural adaptation to acidic environments. In contrast, the significant reduction in growth at pH 8 indicates a sensitivity to alkaline conditions although strain S5 demonstrated relatively higher tolerance under these conditions.

The obtained results are consistent with those reported by Jackson et al. (1991), Kredics et al. (2004), Shafique et al. (2009), Hamdia et al. (2015) and Bao et al. (2024), who found that the optimal growth range for *Trichoderma* lies between pH 4 and 6. However, Cabral-Miramontes et al. (2022) described a broader tolerance in some *Trichoderma* strains, with notable growth up to pH 9, which is not consistent with the obtained results showing a significant reduction in growth from pH 8.

The obtained results also showed an optimal growth of the strains at a temperature of 25 °C and an absence of growth at 35 °C. These findings are similar to the studies of Contreras-Corne-jo et al. (2016), Zehra et al. (2017), Ghazanfar et al. (2018), Andrés et al. (2022), and Meena et al. (2024) who also reported an optimal growth range for *Trichoderma* between 25 °C and 30 °C.

However, significant variability in thermotolerance exists within the *Trichoderma* genus. According to Mukherjee et al. (2012), certain

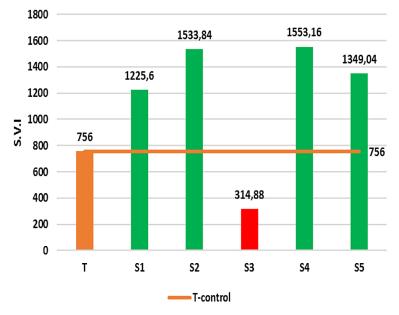


Figure 8. Effect of Trichoderma strains on seed vigor index (S.V.I.)

species, such as *T. pseudokoningii*, exhibit greater temperature tolerance and can grow at higher temperature, while others fail to grow beyond 28 °C. Vinod et al. (2024) demonstrated that *Trichoderma* strains can grow within a temperature range of 15 °C to 35 °C with optimal growth at 30 °C and that some strains can thrive at temperature as high as 40 °C. Furthermore, Poosapati et *al.* (2014) identified *T. asperellum* isolates capable of tolerating high temperature up to 52 °C, illustrating the significant variability in thermotolerance within the *Trichoderma* genus.

Moreover, the result of this study showed that the majority of the tested strains significantly improved the germination rate of tomato seeds compared to the control, with rates ranging from 80% to 86% compared to 70% for the untreated seeds. These results corroborate those obtained by several authors, including Montalvão et al. (2020), Sandri et al. (2023), Vukelic et al. (2024) and Alwadai et al. (2024) who demonstrated that inoculating tomato seeds with Trichoderma promotes germination and enhances root and hypocotyl elongation. Stimulation of germination has been shown to be associated with enhanced nutrient uptake, induction of indole acetic acid (IAA) and secretion of extracellular enzymes (Vukelic et al., 2024). However, the effect of Trichoderma on germination is often isolate specific, and some isolates may inhibit germination (Machado et al., 2015). Santos et al. (2020) reported a reduction in germination rates for certain seeds treated with Trichoderma, Junges et al. (2016) observed a 35.9% decrease in seedling emergence after applying Trichoderma spp. to angico seeds and Pfordt et al. (2024) reported that the germination rate of maize seeds was severely affected by Trichoderma afroharzianum, with only 22%

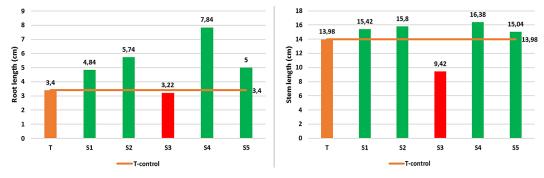


Figure 9. Effect of *Trichoderma* strains on the *in vivo* plant growth promotion assay: a) effect on root length, b) effect on stem length

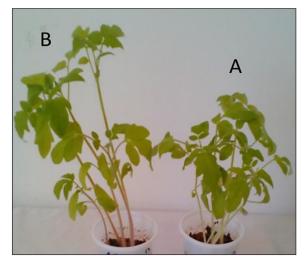


Figure 10. The negative effect of strain S3 on stem length (A) compared to the control (B)

of grains germinating, and the seedlings showed shortened and deformed growth. Furthermore, the application of some commercial *Trichoderma* based products can also have negative effect as demonstrated in the study of Patekoski and Pires-Zottarelli (2010) who observed a decrease in root length in lettuce treated with Biotrich.

The isolate specific effect of *Trichoderma* was also observed in the conducted study. Strain S4 exhibited the best effect on germination reaching a rate of 86% and an S.V.I. significantly higher than the control. However, strain S3 showed a lower S.V.I. due to reduced hypocotyl and root growth.

The analysis of vegetative growth of tomato plant treated with different *Trichoderma* strains showed that strains S1, S2, S4 and S5 exhibited a beneficial effect with a significant increase in root and stem length compared to the control plants, confirming their potential as growth promoters. These results are consistent with those of Uddin et al. (2018), Montalvão et al. (2020), Mahboubi et al. (2023), Alwadai et al. (2024), Leuratti et al. (2025) and Puja et al. (2025), who reported that several *Trichoderma* strains promote root and shoot growth in tomato plants, mainly by stimulating root development and enhancing nutrient uptake.

The production of phytohormones (auxin and gibberellin) and phytoregulators, including the 1-aminocyclopropane-1-carboxylic acid (ACC) deaminase enzyme, is one of the direct mechanisms contributing to the promoting effect of *Trichoderma* on plant growth (Ozimek et al., 2018 ; Jaroszuk-Scisel et al., 2019). Phytohormones participate in the regulation of complex and interrelated plant immune signaling pathways, ensuring a rapid defense response and adaptation to various environmental conditions (Alfiky and Weisskopf, 2021). Morover, *Trichoderma* enhance plant growth by the production of vitamins, solubilization of nutrients present in the soil such asPO<sub>4</sub><sup>3-</sup>, Fe<sup>3+</sup>, Cu<sup>2+</sup>, Mn<sup>4+</sup>, ZnO, and production of siderophores to metal chelation (such as iron, copper, zinc, or magnesium) (Jaroszuk-Scisel et al., 2019 ; Sood et al., 2020 ; Tyskiewicz et al., 2022).

However, notable differences between strains were observed in the conducted study once again highlighting the isolate specific nature of *Trichoderma*'s effect. Strain S3 had a negative impact on stem growth, reducing it to 9.42 cm compared to 13.98 cm in the control. This result aligns with those of Harman et al. (2004) and Montalvão et al. (2020), who reported that certain *Trichoderma* strains had a neutral or even negative effect on plant vegetative growth, emphasizing the importance of selecting strains based on their compatibility with the host plant.

Although widely considered beneficial, some members of the genus *Trichoderma* can also act as pathogens, causing damage and losses of agricultural yield (Pozo et al., 2024). As an example, *Trichoderma virens* was found to be the cause of the death of about six thousand *Polygonatum cyrtonema* plants in China (Gong et al., 2024), *Trichoderma asperellum* is the causative agent of green mold on sweet potato (Yang et al., 2021), *Trichoderma longibrachiatum* has the ability to infect the roots of red-leaf lettuce (Sazali et *al.*, 2023), as well as of *Trichoderma viride* on tomato, pepper, cucumber and pine (Menzies, 1993; Li Destri Nicosia et al., 2015).

This pathogenicity of *Trichoderma* can be explained by its evolutionary origin: mycoparasitism would be the ancestral life form of this fungus, which would then have evolved towards symbiosis due to the interaction with root exudates and phytopathogenic fungi present in the rhizosphere. However, in the absence of adequate immune responses in the plant, particularly those mediated by salicylic acid (SA), *Trichoderma* can excessively colonize roots and become pathogenic. Studies have shown that in SA-deficient model plants, *Trichoderma harzianum* caused systemic rots. Thus, the ability of the plant to activate an SA-based defense response is crucial to maintain the beneficial interaction with *Trichoderma* and prevent pathogenic behavior (Pozo et al., 2024).

In the conducted study, strain S3 decreased stem length without causing any visual lesions or damage to the tomato plant. According to Santos et al. (2020), the effect of *Trichoderma* strains on plant development can vary and this difference depend on the type of crop, inoculation doses, formulations used, and specific environmental conditions. These factors are likely to explain the variable performance of isolates in terms of plant growth promotion.

## CONCLUSIONS

This study highlighted the significant potential of *Trichoderma* strains isolated from lichens as plant growth promoting agents. The results obtained demonstrate the adaptability of these strains to different environmental conditions, in particular their preference for acidic conditions (pH 4.5) and an optimum temperature of 25 °C.

The evaluation of the impact of these strains on the growth of tomato plants revealed promising results, especially for strain S4 and S2. These strains demonstrated their ability to significantly improve not only seed germination, but also the vegetative development of plant. However, the variability of the responses observed between the different strains, illustrated in particular by the negative effects of strain S3, underlines the crucial importance of a rigorous selection of isolates according to their compatibility with host plant. Further research in this area could contribute significantly to improving sustainable agricultural practices.

#### REFERENCES

- Abdul-baki, A.A., Anderson, J.D. (1973). Vigor determination in soybean seed by multiple criteria. Crop. *Science*, 13, 630–633. http://dx.doi.org/10.2135/ cropsci1973.0011183X001300060013x
- Alfiky, A., Weisskopf, L. (2021). Deciphering Trichoderma-plant-pathogen interactions for better development of biological applications. *Journal of Fungi*, 18;7(1):61. https://doi.org/10.3390/ jof7010061
- Alwadai, A.S., Wahibi, M.S., Alsayed, M.F., Alshaikh, N. A., Perveen, K., Elsayim, R. (2024). Molecular characterization of plant

growth-promoting *Trichoderma* from Saudi Arabia. *Scientific Reports* 14, 23236. https://doi. org/10.1038/s41598-024-73762-5

- Andrés, P., Alejandra, P., Benedicto, M., Nahuel, R., Clara, B. (2022). A comparative study of different strains of *Trichoderma* under different conditions of temperature and pH for the control of *Rhizoctonia solani*. *Agricultural Sciences*, *13*, 702–714. https:// doi.org/10.4236/as.2022.136046
- Bao, W., Shen L., Xia, S., Yang, X. (2024). Effect of pH on the growth and competition of *Trichoderma* spp. and *Fusarium* spp. *Chinese Journal of Applied Ecology*, 35(9), 2535–2542. https://doi.org /10.13287/j.1001-9332.202409.032
- Barnett, H.L., Hunter, B.B. (1998). Illustrated genera of imperfect fungi. Fourth edition. Minnesota, USA.
- Cabral-Miramontes, J.P., Olmedo-Monfil, V., Lara-Banda, M., Zúñiga-Romo, E.R., Aréchiga-Carvajal, E.T. (2022). Promotion of plant growth in arid zones by Selected *Trichoderma* spp. strains with adaptation plasticity to alkaline pH. *Biology*. 11, 1206. https://doi.org/10.3390/biology11081206
- Contreras-Cornejo, H.A., Macias-Rodriguez, L., del-Val, E., Larsen, J. (2016). Ecological functions of *Trichoderma* spp. and their secondary metabolites in the rhizosphere: interactions with plants. *FEMS Microbiology Ecology*. 92(4). https://doi. org/10.1093/femsec/fiw036
- Ferreira, F. V., Musumeci, M. A. (2021). *Trichoderma* as biological control agent: scope and prospects to improve efficacy. *World Journal of Microbiology and Biotechnology*, 37(5). https://doi.org/10.1007/ s11274-021-03058-7
- 10. Ghazanfar, U.M., Raza, M., Raza, W. (2018). Effect of physiological parameters on mass production of *Trichoderma* species. *Pakistan Journal of Phytopathology*. 30(1) 59–65. https://doi.org/10.33866/ phytopathol.030.01.0447
- 11. Gong, Z., Yang, Y., Zhang, L., Wang, S., Luo, J., Luo, Q. (2024). First report of Polygonatum cyrtonema root rot caused by Trichoderma virens in China. *Plant disease*, 108: 525. https://doi.org/10.1094/ PDIS-08-23-1647-PDN
- 12. Guzmán-Guzmán, P., Kumar, A., de los Santos-Villalobos, S., Parra-Cota, F.I., Orozco-Mosqueda, M.d.C., Fadiji, A.E., Hyder, S., Babalola, O.O., Santoyo, G. (2023). Trichoderma species: Our best fungal allies in the biocontrol of plant diseases a review. *Plants*, *12*, 432. https://doi.org/10.3390/ plants12030432
- Hamdia, Z., A., Hadi M. A., Naeem S. D., Nibal K. M., Fatimah H. G. (2015). Effects of pH and ECW on growth and sporulation of indigenous *Tricoderma* spp. *International Journal of Phytopathology*. 4(1), 15–20. 10.33687/phytopath.004.01.0966

- 14. Harman, G.E., Petzoldt, R., Comis, A., Chen, J. (2004). Interactions between *Trichoderma harzia-num* strain T22 and maize inbred line Mo17 and effects of these interactions on diseases caused by *Pythium ultimum* and *Colletotrichum graminicola*. *Phytopathology*, 94(2), 147– https://doi. org/153.10.1094/PHYTO.2004.94.2.147
- 15. Jackson, A. M., Whipps, J. M., Lynch, J. M. (1991). Effects of temperature, pH and water potential on growth of four fungi with disease biocontrol potential. *World Journal of Microbiology and Biotechnology*. 7(4): 494–501. https://doi.org/10.1007/ BF00303376
- 16. Jaroszuk-'Sciseł, J., Tyskiewicz, R., Nowak, A., Ozimek, E., Majewska, M., Hanaka, A., Tyskiewicz, K., Pawlik, A., Janusz, G. (2019). Phytohormones (auxin, gibberellin) and ACC deaminase in vitro synthesized by the mycoparasitic Trichoderma DEMTkZ3A0 strain and changes in the level of auxin and plant resistance markers in wheat seedlings inoculated with this strain conidia. *International Journal of Molecular Sciences*, 4; 20(19): 4923. https://doi.org/10.3390/ijms20194923.
- 17. Junges, E., Muniz, M.F., Mezzomo, R., Bastos, B., Machado, R.T. (2016). *Trichoderma* spp. in the production of seedling of forest species. *Floresta e Ambiente*. 23(2), 237–244. https://doi. org/10.1590/2179-8087.107614
- 18. Kredics, L., Manczinger, L., Antal, Z., Penzes, Z., Szekeres, K., Kevel, F., Nagy, E. (2004). In vitro water activity and pH dependence of mycelial growth and extracellular enzyme activities of *Trichoderma* strains with biocontrol potential. *Journal of Applied Microbiology*. 96(3): 491–8. https://doi. org/10.1111/j.1365-2672.2004.02167.x
- 19. Kredics, L., Büchner, R., Balázs, D., Allaga, H., Kedves, O., Racić, G., Varga, A., Nagy, V.D., Vágvölgyi, C., Sipos, G. (2024). Recent advances in the use of Trichoderma-containing multicomponent microbial inoculants for pathogen control and plant growth promotion. *World Journal of Microbiology* and Biotechnology, 40, 162.https://doi.org/10.1007/ s11274-024-03965-5
- 20. Leuratti, T., Fellin, L., Michelon, N., Palacios Tario, J.B., Gutiérrez, J.E.S., Gianquinto, G., et al. (2025). Optimizing tomato seedling production in the tropics: Effects of Trichoderma, Arbuscular Mycorrhizal Fungi, and key agronomical factors. Agronomy, 15(2), 1–20. https://doi.org/10.3390/ agronomy15020392
- 21. Li Destri Nicosia, M.G., Mosca, S., Mercurio, R., Schena, L. (2015). Dieback of Pinus nigra seedlings caused by a strain of Trichoderma viride. *Plant Disease*, 99: 44–49. https://doi.org/10.1094/ PDIS-04-14-0433-RE
- 22. Machado, D.F.M., Tavares, A.P., Lopes, S.J.,

Silva, A.C.F. (2015). *Trichoderma* spp. In emergence and growth of cambará seedlings (*Gochnatia polymorpha* (Less.) Cabrera). *Revista Árvore*. *39*(1): 167–176. http://dx.doi. org/10.1590/0100-67622015000100016

- 23. Mahboubi, S., Bouchikh, Y., Latreche, A., Da Lio, D., Chamekh, R. (2023). Biocontrol and growth promoting effect of *Trichoderma* spp. isolate from rhizosphere soils of some Algerian areas. *South Asian Journal of Experimental Biology*, *13*, 52–61. https:// doi.org/10.38150/sajeb.13(1).p52-61
- 24. Meena, N., Yadav, D.L., Gautam, C., Yadav, V.K., Yadav, S. L., Meena, C. B. (2024). Trichoderma isolates against abiotic stresses and management of collar rot of lentil (*Lens culinaris* L.) caused by sclerotium rolfsii. *Indian Journal of Microbiology*. 64, 1366–1375. https://doi.org/10.1007/ s12088-024-01356-w
- 25. Mellab, K. (2025). Agricultural and agri-food exports: Where does Algeria stand ?. *African Scientific Journal*, 3(28), 2658–9311. https://doi.org/10.5281/zenodo.15075401
- 26. Menadi, S., Boubidi, F.S., Bouchelaghem, R. et al. (2024). Occupational exposure to the dust of chemical fertilizers (NPK 15.15.15): effect on biochemical parameters and oxidative stress status among workers in Annaba. *Comparative Clinical Pathology. 33*, 437–444. https://doi.org/10.1007/ s00580-024-03563-9
- 27. Menzies, J.G. (1993). A strain of Trichoderma viride pathogenic to germinating seedlings of cucumber, pepper and tomato. *Plant Pathology*, 42: 784–791. https://doi.org/10.1111/j.1365-3059.1993.tb01565.x
- 28. Montalvão, S. C. L., Marques, E., Silva, J. B. T., Silva, J. P., Mello, S. C. M. (2020).*Trichoderma* activity in seed germination, promoting seedling growth and Rhizocompetence in tomato plants. *Journal of Agricultural Science*. 12(10): 252–262. https://doi.org/10.5539/jas.v12n10p252
- Mukherjee, M., Mukherjee, P.K., Horwitz, B.A., Zachow, C., Berg, G., Zeilinger, S. (2012). Trichoderma-plant-pathogen interactions: Advances in genetics of biological control. *Indian Journal of Microbiology*. *52*, 522–529. https://doi.org/10.1007/ s12088-012-0308-5
- 30. Oh, S.Y., Yang, J.H., Woo, J.J., Oh, S.O., Hur, J.S. (2020). Diversity and distribution patterns of Endolichenic Fungi in Jeju Island, South Korea, *Sustainability*. 12, 3769. https://doi.org/10.3390/ su12093769
- 31. Ozimek, E., Jaroszuk-'Sciseł, J., Bohacz, J., Korniłłowicz-Kowalska, T., Ty'skiewicz, R., Słomka, A., Nowak, A., Hanaka, A. (2018). Synthesis of indole acetic acid, gibberellic acid and ACCdeaminase by Mortierella strains promote winter wheat seedlings growth under different conditions.

International Journal of Molecular Science., 19, 3218. https://doi.org/10.3390/ijms19103218

- 32. Patekoski, K.S., Pires-Zottarelli, C.A. (2010). Pythium aphanidermatum pathogenicity to hydroponics lettuce and its biocontrol with Trichoderma. *Pesquisa Agropecuária Brasileira*. 45(8), 805–810. https:// doi.org/10.1590/s0100-204x2010000800005
- 33. Pfordt, A., Steffens, L.Ä., Raz, T., Naumann, M. (2024) Impact of Trichoderma afroharzianum infection on fresh matter content and grain quality in maize. *Frontier Plant Science*. 15: 1436201. https:// doi.org/10.3389/fpls.2024.1436201
- 34. Pfordt, A., Douanla-Meli,C., Schäfer, B.C., Schrader, G., Tannen, E., Chandarana, M.J., von Tiedemann, A. (2025). Phylogenetic analysis of plant-pathogenic and non-pathogenic *Trichoderma* isolates on maize from plants, soil, and commercial bio-products. *Applied and Environmental Microbiology*. 19; 91(3):e0193124. https://doi. org/10.1128/aem.01931-24
- 35. Poosapati, S., Ravulapalli, P.D., Tippirishetty, N., Vishwanathaswamy, D.K., Chunduri, S. (2014). Selection of high temperature and salinity tolerant Trichoderma isolates with antagonistic activity against *Sclerotium rolfsii*. *Springer Plus.* 3, 641. https://doi.org/10.1186/2193-1801-3-641
- 36. Pozo, M.I., Herrero, B., Martín-García, J., Santamaría, Ó., Poveda, J. (2024) Evaluating potential side effects of Trichoderma as biocontrol agent: A two-edges sword?. *Current Opinion in Environmental Science & Health*: 41: 100566. https://doi. org/10.1016/j.coesh.2024.100566
- 37. Puja, J., Ram Bahadur, K., Suraj, B., Aashaq, H.B., Arvind, K. K. (2025). Nematicidal efficacy of native *Trichoderma isolates* on Meloidogyne incognita and their influence on tomato growth parameters. *Journal of Natural Pesticide Research. 12*, 100128. https://doi.org/10.1016/j.napere.2025.100128
- Rubio, M.B., Domínguez, S., Monte, E., Hermosa, R. (2012). Comparative study of *Trichoderma* gene expression in interactions with tomato plants using high-density oligonucleotide microarrays. *Microbiology*, *158*, 119–128. 10.1099/mic.0.052118-0. Epub 2011 Nov 10.
- 39. Sandri, M.R., Cavião, H.C., Oliveira, C.F., Andrade, L.B., Granada, C., Schwambach, J. (2023). The plant growth effect and biocontrol potential of *Trichoderma* sp. inoculation in tomatoes are dependent of the inoculation way. *Bragantia*, 83, 13. https://doi.org/10.1590/1678-4499.20230075
- 40. Santos, M. F. D., Santos, L. E. D., Costa, D. L. D., Vieira, T. A., Lustosa, D. C. (2020). *Tricho-derma* spp. on treatment of *Handroanthus ser-ratifolius* seeds: effect on seedling germination and

development.*Heliyon*, 6(6), e04044. https://doi. org/10.1016/j.heliyon.2020.e04044

- 41. Sazali, M., Zakry, F.A.A., Kundat, F.R. (2023). Plants wilt disease of red leaf lettuce (Lactuca sativa L.) after colonized by Trichoderma longibrachiatum. *Malaysian Applied Biology*, 52, 163–176. https://doi.org/10.55230/mabjournal.v52i5.icfic12
- Shafique, S., Bajwa, R., Shafique, S. (2009) Cellulase biosynthesis by selected *Trichoderma* species. *Pakistan Journal of Botany*. 41(2): 907–916.
- 43. Sood, M., Kapoor, D., Kumar, V., Sheteiwy, M.S., Ramakrishnan, M., Landi, M., Araniti, F., Sharma, A. (2020). Trichoderma: The "secrets" of a multitalented biocontrol agent. *Plants*, *9*, 762. https://doi. org/10.3390/plants9060762
- 44. Tucci, M., Ruocco, M., de Masi, L., de Palma, M., Lorito, M. (2011). The beneficial effect of *Trichoderma* spp. on tomato is modulated by the plant genotype. *Molecular Plant Pathology*. *12*(4), 341–354. 10.1111/j.1364-3703.2010.00674.x
- 45. Tyskiewicz, R., Nowak, A., Ozimek, E., Jaroszuk-'Sciseł, J. (2022). Trichoderma: The current status of its a sood pplication in agriculture for the biocontrol of fungal phytopathogens and stimulation of plant growth. *International Journal of Molecular Sciences*. 23, 2329. https://doi.org/10.3390/ ijms23042329
- 46. Uddin, M. N., Rahman, U., Khan, W., Uddin, N., Muhammad, M. (2018). Effect of *Trichoderma harzianum* on tomato plant growth and its antagonistic activity against *Pythium ultimum* and *Phytophthora capsici. Egyptian Journal of Biological Pest Control.* 28, 32. https://doi.org/10.1186/s41938-018-0032-5
- 47. Vinod, C., Veena, S.S., Sreekumar, J., Karthikeyan, S., Jeeva, M.L. (2024). Assessment of variability in temperature tolerance and antagonistic activity among Trichoderma isolates for biological control applications. *Journal of Root Crops.* 50(1), 23–31.
- Vukelic, I., Radic, D., Pecinar, I., Levic, S., Djikanovic, D., Radotic, K., Pankovic, D. (2024). Spectroscopic Investigation of Tomato Seed Germination Stimulated by Trichoderma spp. *Biology*, 13, 340. https://doi.org/10.3390/biology13050340
- 49. Yang, Y., Fang, B., Feng, S., Wang, Z., Luo, Z., Yao, Z., et al. (2021). Isolation and identification of Trichoderma asperellum, the novel causal agent of green mold disease in sweetpotato. *Plant Disease*, 105: 1711–1718. https://doi.org/10.1094/ PDIS-07-20-1484-RE
- 50. Zehra, A., Dubey, M.K., Meena, M., Upadhyay, R.S. (2017). Effect of different environmental conditions on growth and sporulation of some Trichoderma species. *Journal of Environmental Biology*. 38, 197– 203. https://doi.org/10.22438/jeb/38/2/MS-251