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Article

Influence of *Sargassum* spp. extract on germination and antioxidant activity of tomato seedlings

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Abstract

In recent years, the macroalga *Sargassum* spp. has become an economic and environmental problem in several coastal regions. Nonetheless, due to the wide variety of bioactive compounds it possesses, it has been proposed as a sustainable alternative in agriculture, as it can improve seed germination, nutrient absorption, and photosynthesis, and mitigate biotic and abiotic stress. The objective of this study was to evaluate the effect of priming tomato seeds with aqueous extracts of *Sargassum* spp. on germination, vigor, biomass, photosynthetic pigments and some indicators of the antioxidant system of seedlings. The concentrations of the extracts were: 0.5, 1.5, 2.5 and 3.5%, and a control with distilled water. The results indicate that the extracts improved germination, vigor, phenolic compounds, flavonoids, antioxidant capacity and photosynthetic pigments. There were no significant improvements in biomass. These results suggest that applying *Sargassum* spp. extracts may be an ecological alternative to improve germination parameters and stimulate the synthesis of secondary metabolites in tomato seedlings.

Keywords :

Solanum lycopersicum L., biostimulants, seed priming.

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Introduction

Modern agriculture faces the challenge of increasing crop productivity and quality under criteria of sustainability and food security (FAO, 2025). The growing demand for food drives strategies that optimize agricultural yield while reducing environmental impact. In this context, natural biostimulants stand out for their capacity to improve production and quality, decreasing the use of agrochemicals and mitigating biotic and abiotic stress (Durán-Hernández *et al*., 2022). Among them, seaweed derivatives, such as *Sargassum* spp., are promising due to their wealth of bioactive compounds. These macroalgae contain polysaccharides, phenolic compounds, carotenoids, phytohormones and amino acids, which improve germination, growth and quality, as well as confer tolerance to salt, water, thermal and pathogen stress (Senthilkumar *et al*., 2024).

Although its massive proliferation generates environmental and economic problems in the Mexican Caribbean, its high content of secondary metabolites makes it a valuable source of agricultural biostimulants (Adderley *et al*., 2023). Previous studies show its potential to improve the growth and nutraceutical quality of different crops (Rivera-Solís *et al*., 2021).

For its part, the tomato (*Solanum lycopersicum* L.) is one of the most cultivated and consumed vegetables worldwide, appreciated for its nutritional and antioxidant value (Collins *et al*., 2022). To ensure the productivity of this crop, high-quality seeds are required to ensure uniform germination, a key stage for crop yield (Ruiz-Ramirez *et al*., 2021; Reed *et al*., 2022). Germination, the transition from dormancy to active growth, can be optimized by pre-germination treatments such as priming, which improves the germination rate, vigor and stress tolerance (Wang and Shi, 2024; Abinandan *et al*., 2025). Given the biostimulant potential of *Sargassum* spp., this study evaluated the effect of *Sargassum* spp. extracts on germination, photosynthetic pigments and antioxidant activity in tomato seedlings.

Materials and methods

Liquid extract of *Sargassum* spp.

Brown algae *Sargassum* spp., previously dehydrated and pulverized, provided by the Antonio Narro Agrarian Autonomous University (UAAAN), was used. The liquid extract was prepared according to the methodology of Sariñana-Aldaco *et al*. (2021), where its biochemical characterization is detailed. A mother solution (100%) was prepared in a 1:20 ratio (m:v, algae:distilled water), boiled for 1 h, pressed and filtered. From this concentrated solution, the required dilutions (0.5, 1.5, 2.5 and 3.5%) were obtained with bidistilled water.

Germination test

Saladette tomato seeds of the 'Río Grande' variety were used, disinfected with 75% ethanol for 5 min and washed twice with distilled water. The seeds were soaked for 24 h in the concentrations of the *Sargassum* spp. extract (0.5, 1.5, 2.5 and 3.5%), including a control with distilled water. Subsequently, they were placed on #1 filter paper inside sterile Petri dishes (90 mm), with 10 seeds per dish and 20 dishes per treatment. The filter paper was moistened with sterile distilled water. The experimental design was completely randomized with five treatments and 20 repetitions. The Petri dishes were incubated in a germination chamber (Achieva, Lab-Tech Inc, Model No. A-3920), under a 12-hour day/night cycle, at 25 ±2 °C and 60% relative humidity (Li *et al*., 2019). After 14 days, the germination percentage, fresh biomass and vigor percentage were evaluated.

Germination percentage (G) and vigor (V) were calculated using the following formulas:

1)

$$G = \frac{n}{N} \times 100$$

Where: n= number of germinated seeds on day 14; N= total number of seeds.

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2)

$$V = \frac{Pn}{N} \times 100$$

Where: Pn= normal seedlings; N= total number of seeds.

Photosynthetic pigments

Chlorophyll content was determined according to the method of Lichtenthaler and Wellburn (1983), using 95% ethanol as a solvent. Absorbance readings were performed at 665, 649, and 470 nm in a UV-visible spectrophotometer (Jenway 7305).

Chlorophyll concentrations were calculated using the following equations:

3)

$$Chlorophylla = 13.95A_{665} - 6.88A_{649}$$

4)

$$Chlorophyllb = 24.96A_{649} - 7.32A_{665}$$

5)

$$Totalchlorophyll = chlorophylla + chlorophyllb$$

Extraction of bioactive compounds

A quantity of 2 g of fresh sample was homogenized with 10 ml of 80% ethanol, by constant orbital agitation (70 rpm) for 24 h at 70 rpm and 5 °C. Subsequently, the extracts were centrifuged at 3 000 rpm for 5 min, and the supernatant was used for subsequent analyses.

Bioactive compounds

The content of phenolic compounds was determined using an adaptation of the Folin-Ciocalteu method (Singleton *et al*., 1999). Fifty µl of ethanolic extract was mixed with 3 ml of distilled water and 250 µl of Folin-Ciocalteu reagent (1 N). After 3 minutes of rest, 750 µl of Na_2CO_3 and 950 µl of distilled water were added, incubating for 2 h. Absorbance was measured at 765 nm in a UV-visible spectrophotometer (Jenway 7305), expressing the results as mg of gallic acid equivalents (GA) per 100 g of fresh weight (FW).

Total flavonoid content was quantified using the colorimetric method described by Buendía-García *et al*. (2021). 250 µl of extract were mixed with 1.25 ml of distilled water and 75 µl of $NaNO_2$ (5%), resting for 5 min. Then, 150 µl of $AlCl_3$, 500 µl of NaOH (1 M), and 275 µl of distilled water were added. Absorbance was measured at 510 nm, and the results were expressed in mg of quercetin equivalents (QE) per 100 g FW.

Antioxidant capacity was evaluated using the DPPH+ method (Brand-Williams *et al*., 1995), using a solution of DPPH+ in ethanol (0.025 mg ml^{-1}). A quantity of 50 µl was mixed with 1 950 µl of DPPH +, incubated for 30 min, and absorbance was measured at 517 nm. The results were expressed in milliequivalents (Meq) of Trolox per 100 g of FW.

Catalase activity

Catalase activity (CAT, EC 1.11.1.6) was determined according to the method of Aebi (1974). Readings were performed at room temperature in a UV-visible spectrophotometer (Jenway 7305), monitoring the decrease in absorbance at 240 nm due to H_2O_2 decomposition. Enzymatic activity was calculated using the molar extinction coefficient ($\#_{240}$ = 43.6 M cm^{-1}) and protein concentration, determined by the method of Bradford (1976). Results were expressed as U mg^{-1} protein, defining one unit (U) as the amount of enzyme that causes a 0.001 absorbance change per minute under the assay conditions.

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Statistical analysis

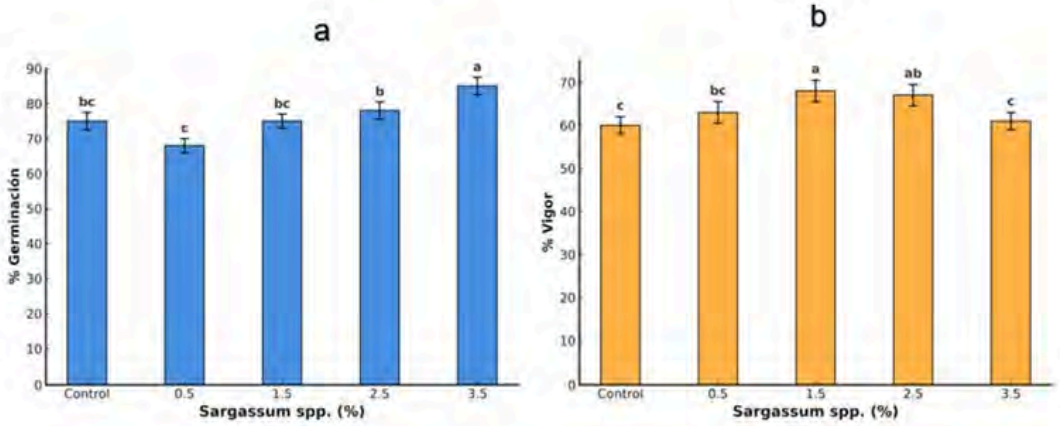
Data normality was verified using the Kolmogorov-Smirnov test. Data expressed as a percentage were transformed using arcsine or square root functions, depending on their distribution. The analysis of variance and the comparison of means by Tukey's test ($p \leq 0.05$) were performed with the Statistical Analysis System (SAS) statistical package, version 9.3.

Results and discussion

Germination test

Germination and vigor are critical stages in plant development, determining crop establishment and yield (Papoui *et al*., 2025). In this study, treatment with the liquid extract of *Sargassum* spp. significantly improved the germination and vigor of tomato seeds (Figure 1). The 3.5% concentration increased germination by 19%, while 1.5% increased vigor by 13% compared to the control.

Figure 1. Effect of *Sargassum* spp. liquid extract on germination (a) and vigor (b) of tomato seeds. Different letters indicate significant differences between treatments (Tukey, $p \leq 0.05$).



Similar results have been reported in tomatoes with brown algae extracts (Hernández-Herrera *et al*., 2023) and in *Abelmoschus esculentus* with *Ecklonia maxima* extracts (Makhaye *et al*., 2021). The positive effect is attributed to the presence of phytohormones, polysaccharides and antioxidant compounds that regulate key processes during germination (Cossa *et al*., 2023; Margal *et al*., 2023). Brown algae, which contain auxins, gibberellins and cytokinins that facilitate the breaking of dormancy by activating hydrolytic enzymes, such as amylase, which converts starch into simple sugars for embryonic development (Sariñana-Aldaco *et al*., 2022). In addition, these phytohormones promote cell division and elongation (Zluhan-Martínez *et al*., 2021). Nevertheless, its effectiveness depends on multiple factors, such as the species of algae, type of extraction, environmental conditions, concentration and the receiving plant species (Martínez-González *et al*., 2022). High concentrations can cause adverse effects due to the accumulation of compounds that alter the hormonal balance or induce physiological stress (Castro *et al*., 2022).

Biomass production

Priming with liquid extract of *Sargassum* spp. did not cause significant effects on biomass (Figure 2). These results agree with previous studies that report no differences between different concentrations used. For example, Morales-Meléndez *et al*. (2023) observed no significant increases in biomass production of tomato plants treated with seaweed extracts, and Rivera-

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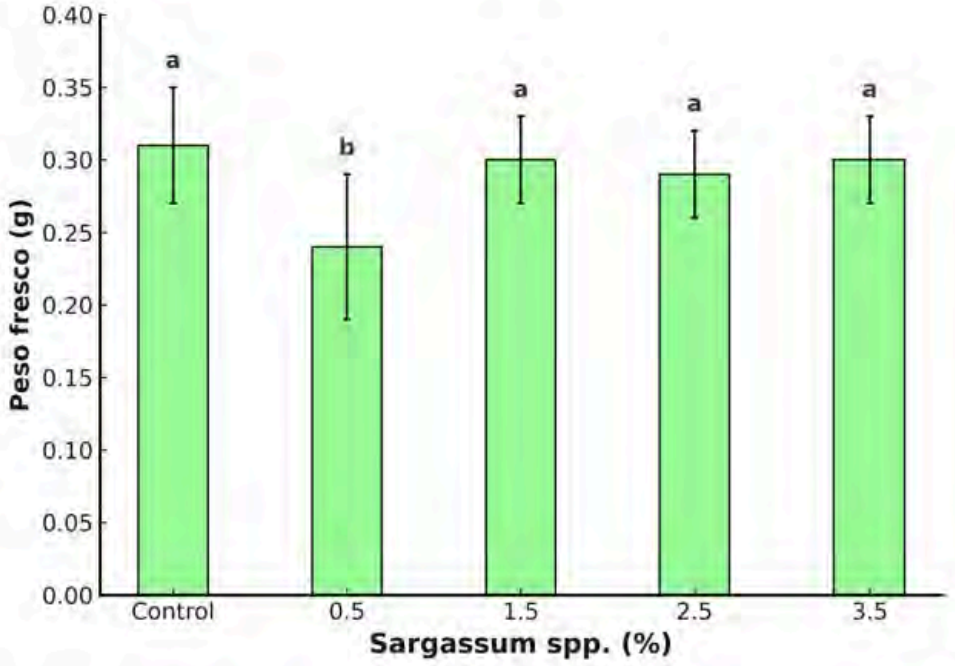
Solís *et al*. (2021) found no differences compared to the control. This suggests that the chemical composition of the extracts and their concentration are key factors in inducing a positive physiological response or preventing inhibitory effects.

Figure 2. Fresh weight of tomato seedlings treated with *Sargassum* spp. extract. Different letters indicate significant differences between treatments (Tukey, $p \leq 0.05$).



Photosynthetic pigments

Total chlorophyll and carotenoids are key indicators of the photosynthetic capacity and physiological state of plants (Sherin *et al*., 2022). In this study, both pigments were significantly affected by *Sargassum* spp. extracts (Table 1). The 3.5% concentration reduced total chlorophyll by 14.85% compared to the control. Similar results have been reported with seaweed extracts (Salazar-Salazar *et al*., 2022; Mohammed *et al*., 2023).

Table 1. *Sargassum* spp. extract on the content of total chlorophyll and carotenoids in tomato seedlings.

Sargassum spp. extract (%)	Total chlorophyll (mg g^{-1} DW)	Carotenoids (mg g^{-1} DW)
Control	1.28 b	102.68 b
0.5	1.48 ab	118.27 ab
1.5	1.91 a	137.8 a
2.5	1.61 a	128.44 ab
3.5	1.09 b	82.91 b

DW= dry weight. Different letters indicate significant differences between treatments (Tukey, $p \leq 0.05$).

This effect has been attributed to the action of betaines present in the extracts, which act as molecular chaperones stabilizing sensitive biomolecules, such as pigments, and preventing their degradation (Sariñana-Aldaco *et al*., 2025). Such compounds have been shown to prevent the loss of photosynthetic activity by inhibiting chlorophyll degradation (Genard et al., 1991), especially at intermediate doses.

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Bioactive compounds and catalase activity

The plant antioxidant system comprises two components: enzymatic, considered the first line of defense, integrated by enzymes such as catalase (CAT), ascorbate peroxidase (APX) and glutathione peroxidase (GPX), which degrade reactive oxygen species (ROS) such as H_2O_2 (Dumanovi# *et al*., 2021), and non-enzymatic, made up of antioxidant metabolites such as ascorbic acid, phenolic compounds, glutathione and carotenoids (Hashim *et al*., 2020). In this study, the *Sargassum* spp. extract (1.5%) significantly increased total phenols (141%), flavonoids (125%), and antioxidant capacity (11%) compared to the control (Figure 3). However, CAT activity decreased in treatments with 1.5% and 3.5% (Figure 4). These results agree with reports on the positive effect of seaweed extracts on the accumulation of bioactive compounds (Sariñana-Aldaco *et al*., 2021); nevertheless, they differ from studies that report increases in antioxidant enzymes in soybeans treated with *Ascophyllum nodosum* extracts (Repke *et al*., 2022).

Figure 3. Total phenols(a), flavonoids(b) and antioxidant capacity (c) in tomato seedlings treated with liquid extract of *Sargassum* spp. Different letters indicate significant differences between treatments (Tukey, $p \leq 0.05$).

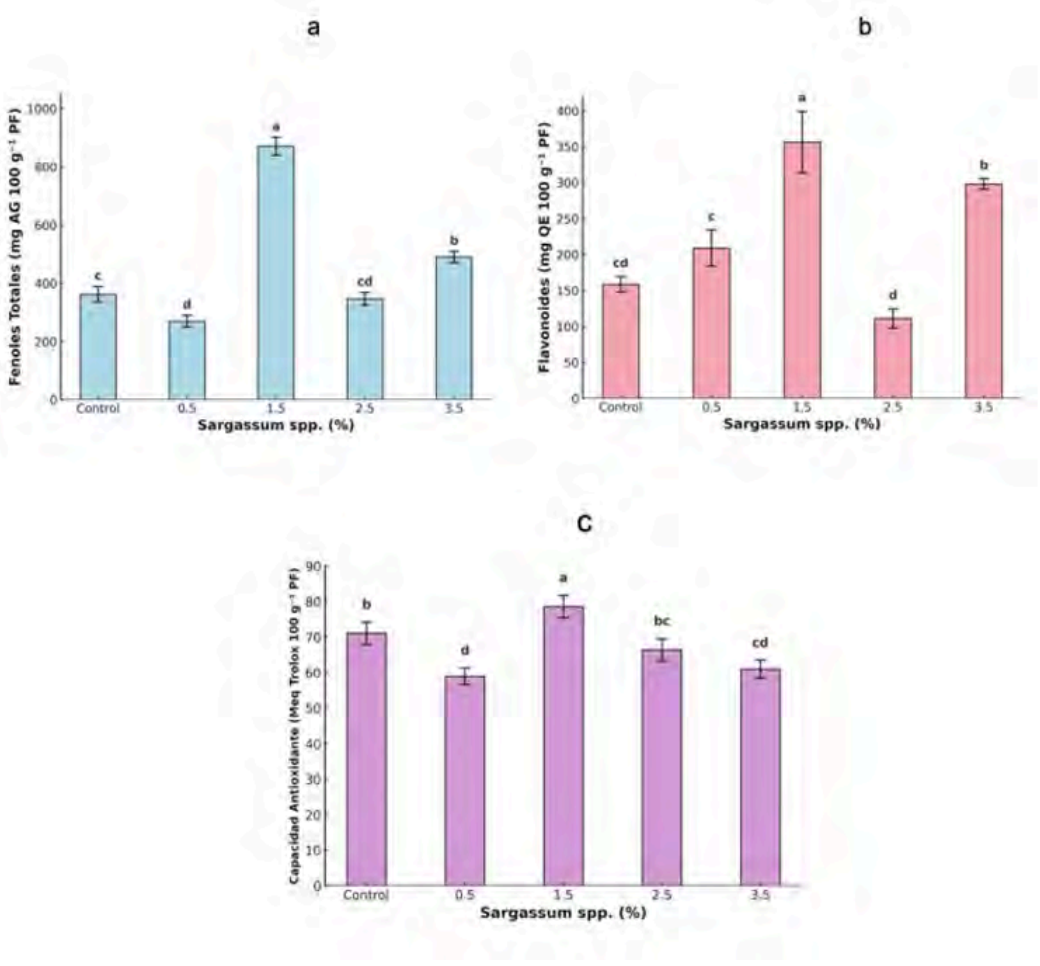

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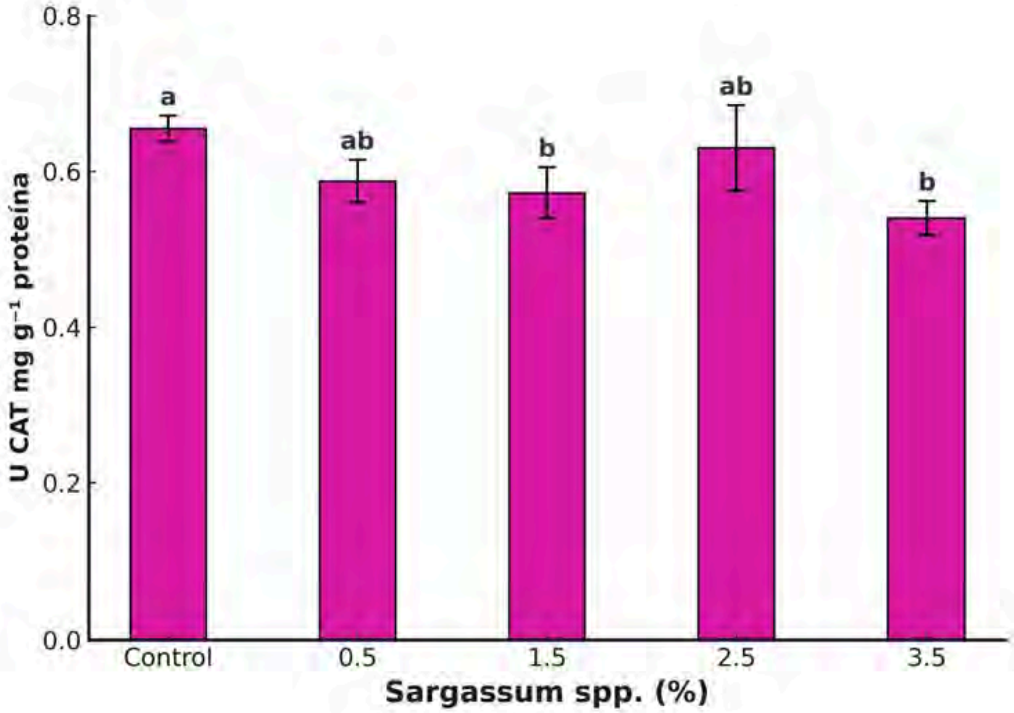
Figure 4. Catalase activity in tomato seedlings treated with *Sargassum* spp. extract. Different letters indicate significant differences between treatments (Tukey, $p \leq 0.05$).



The observed response can be attributed to metabolites of the extract that activate signaling and gene expression cascades for antioxidant enzymes and synthesis of phenolic compounds (González-Morales *et al*., 2021). In addition, some metabolites exert direct action on the cytoplasm (Lau *et al*., 2025). The reduction in CAT suggests a rebalancing of the antioxidant system, favoring non-enzymatic defense through the biosynthesis of phenols and flavonoids, which decreases the need for enzymatic pathways to neutralize ROS (Kumar *et al*., 2024).

Conclusions

Sargassum spp. extract, as a natural biostimulant, improved tomato seed germination, seedling vigor and the biosynthesis of bioactive compounds, thereby increasing their antioxidant capacity. *Sargassum* spp. extract is a sustainable alternative to promote germination, seedling development, and increased production of secondary metabolites.

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