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RESEARCH ARTICLE

Assessment of the effects of inoculation with entomopathogenic fungi on the vegetative growth and yield of *Capsicum chinense* under water stress conditions

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Abstract

W. Chan-Cupul, J. M. Palma-García, E. Ruiz-Sánchez, and E. Cruz-Crespo. 2025. Assessment of the effects of inoculation with entomopathogenic fungi on the vegetative growth and yield of *Capsicum chinense* under water stress conditions. Int. J. Agric. Nat. Resour. 28-39. In Mexico, *Capsicum chinense* has significant economic, cultural, and gastronomic value. The cultivation of *C. chinense* is increasingly challenged by global warming and droughts, which impact both plant health and market stability. Climate change affects agriculture by altering temperature and precipitation patterns, leading to soil moisture loss, drought, phenological damage, and increased pest and disease incidence. The use of bioinoculants, including entomopathogenic fungi (EFs), may be a strategy to mitigate drought in *C. chinense* production. The aim of this study was to assess the impact of *Beauveria brongniartii* and *Purpureocillium lilacinum* inoculation on the vegetative growth and yield of *C. chinense* var. "Chichen Itza" under two water stress conditions. Experiments were conducted in a gothic greenhouse, and *C. chinense* seedlings were transplanted into growth bags [coconut fiber (70%) and dust (30%)] with controlled irrigation at 75% and 100% levels. The EFs were applied weekly for the first month (50 mL at 1×10^{7} conidia mL^{-1}). Agronomic parameters, including plant height, stem diameter, chlorophyll index, leaf area, fruit quality, and yield, were measured. The results indicated that both *B. brongniartii* and *P. lilacinum* significantly increased plant height and stem diameter in the early stages of growth under water stress conditions. *B. brongniartii* notably increased plant growth and maintained fruit yield even under reduced irrigation. However, no significant differences were observed in the chlorophyll index or overall fruit yield among the treatments. The results of this study suggest that *B. brongniartii* and *P. lilacinum* can improve *C. chinense* resilience to water stress, suggesting potential applications in sustainable agriculture amidst climate change.

Keywords: *Beauveria*, coconut fiber, fruit size, habanero pepper, *Purpureocillium*.

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Highlights

- *Beauveria brongniartii* enhances the height and stem diameter of *Capsicum chinense* plants.

- Inoculation with *B. brongniartii* increases the chlorophyll index in *C. chinense* plants.

- A 25% reduction in irrigation decreases the height and stem diameter of *C. chinense* plants.

- Inoculation with entomopathogenic fungi helps *C. chinense* plants maintain their yield under water stress.

- Water stress in *Capsicum chinense* plants decreases fruit weight.

Introduction

The *Capsicum* genus comprises 27 species, with only five being domesticated. *Capsicum chinense* Jacq. is commercially cultivated on a limited scale in various regions across North and Central America, West and Central Africa, and Oceania. *Capsicum chinense f*ruits are used for multiple purposes, such as fresh vegetables (hot peppers), drying spices (paprika and chili powder), and food coloring, flavoring, and other compounds with medicinal or industrial applications (Rai et al., 2023; Keyton et al., 2014).

In Mexico, *C. chinense* has economic, cultural, and gastronomic significance, is consumed fresh for its spiciness and serves as a vital ingredient in the gastronomy of southeastern Mexico, especially in the Yucatan Peninsula, which has a designation of origin (Meneses-Lazo & Garruña, 2020). The cultivation of *C. chinense*, like many other crops, faces challenges from global warming and droughts, which impact both the plants and the international market (Kim et al., 2023). Agriculture, which is highly susceptible to environmental changes in temperature and precipitation, is a primary sector affected by climate change worldwide. In recent decades, climate change has posed significant risks to humanity, particularly in terms of food security and overall development (Das et al., 2020).

The effects of climate change are manifested through atypical variations in climate elements that influence, for example, the availability of water and, in turn, the production of different types of crops (Pereyda-González et al., 2022). Direct impacts on plants include soil moisture loss, drought, phenological damage, erratic flowering, and an increase in pests and diseases, which are attributable to the rising temperature estimated at two degrees Celsius over this century (Srikanth et al., 2019). In response to drought caused by climate change, plants activate defense mechanisms such as photorespiration, leading to decreased photosynthetic efficiency but allowing heat dissipation caused by excess light energy. Over time, plants have evolved responses to constant water deficit, including stomatal closure mediated by abscisic acid (Baslam et al., 2020).

Studies have shown that plant-microorganism associations increase plant resistance to biotic and abiotic stress through the production of secondary metabolites, including phytohormones (auxins, gibberellins, cytokinins, and abscisic acid) and antifungal-antibacterial compounds (organic acids, siderophores, and enzymes) (Baron et al., 2020). Notably, Toscano-Verduzco et al. (2019) reported the ability of a native strain of *Beauveria brongniartii* (Sacc.) Petch. to solubilize inorganic phosphates [$Ca_3(PO_4)_2$ and $FePO_4$] associated with *C. chinense* plant growth. More recently, Baron et al. (2020) demonstrated that *Purpureocillium lilacinum* (Thom) Luangsaard, Houbraken, Hywel-Jones & Samson and *Metarhizium marquandii* (Massee) Kepler, S.A. Rehner & Humber reduce abiotic stress in maize (*Zea mays* L.), bean (*Phaseolus vulgaris* L.), and soybean [*Glycine max* (L.) Merr.], introducing a novel application of entomopathogenic fungi in biological-organic farming.

On the other hand, some studies have reported the ability of *B. brongniartii* and *P. lilacinum* to improve the growth of *C. chinense*. These studies show that entomopathogenic fungi also improve plant development by colonizing plant tissues, increasing the resistance of plants to pest insects (Toscano-Verduzco et al., 2019; Moreno-Salazar et al., 2020). Although some studies have highlighted the growth-promoting effects of *B. brongniartii* and *P. lilacinum* on *C. chinense*, their potential as biofertilizers against water stress remains unexplored. Therefore, the aim of this study was to assess the impact of *B. brongniartii* and *P. lilacinum* inoculation on the vegetative growth and yield of *C. chinense* var. "Chichen Itza" under two water stress conditions.

Materials and methods

Experimental site

The study was carried out in a gothic greenhouse (20×10 threads cm^2) situated in the Faculty of Biological and Agricultural Science of the University of Colima in Tecoman, Colima, Mexico (18°57'09" N -103°53'45" W). The climate in the region is warm subhumid A(w0) according to the Köppen-García climate classification, with an average temperature of 26.3 °C, 750 mm of precipitation per year, and an altitude of 33 m.

Biological material

Capsicum chinense seeds var. "Chichen Itza" (Bayer® Crop Science) were used as the plant material. Both *B. brongniartii* and *P. lilacinum* were isolated from an organic papaya (*Carica papaya* L.) orchard in Tecoman, Colima, Mexico, and identified morphologically and molecularly by Chan-Cupul et al. (2014).

Fungal conidia production

B. brongniartii and *P. lilacinum* conidia were produced at high concentrations using the solid-state fermentation process in rice grains (*Oryza sativa* L.). Whole grains of rice were washed three times to prevent contaminants. The first wash was carried out with drinking water to remove dust and impurities, the second wash was carried out with a solution of Full-Gro® (quaternary ammonium salt, 1 m L^{-1}) in water with a rest time of 15 minutes, and finally, the last wash was carried out with drinking water to eliminate the disinfectant residue. The clean rice was dried in the sun for 25 minutes, and 120 g of clean and dry rice was placed in high-density polyethylene bags with a capacity of two kilograms. The bags with rice were sterilized at 120 °C for 20 minutes in an autoclave, and the bags were allowed to cool and rest for 24 h. In a laminar flow chamber, the bags were inoculated with 10 mL of a conidial mixture at a concentration of 1×10^7 conidia mL^{-1}, and each EF species was inoculated separately.

The conidial mixture solution was made from a Petri dish of each 18-day-old fungus, and 10 mL of distilled water with Tween 80 (0.05%) was poured onto the surface of the colony and scraped with a sterile spatula, then filtered into a Falcon tube (50 mL) through sterile gauze after it was adjusted to 250 mL with sterile distilled water containing Tween 80 (0.05%). The inoculated bags were incubated at 25±3.0 °C, 80±5.0% relative humidity, and 14:10 h light: dark cycle for 16 days. Every third day, the bags were gently moved to prevent compaction of the substrate due to mycelial growth and to allow aeration and sporulation of the HFs. The conidia were harvested by shaking in a PVC cylinder (30 cm high × 16 cm diameter) with 40 µm mesh lids. For each bag of substrate, 5 g of micronized diatoms (5-10 µm) was used as a carrier. The concentration of conidia/g was determined in a Neubauer chamber. The resulting powder (conidia + carrier) was used as an active agent (Toscano-Verduzco et al., 2019).

Capsicum chinense seedlings and transplanting

Seeds were planted in disinfected (FullGro® 1 mL L^{-1}) polyethylene trays (200 cavities). The trays were filled with *Sphagnum* (Peat Moss®, 4 kg $tray^{-1}$). One *C. chinense* seed was placed in each cavity, approximately one centimeter deep. The trays were completely covered with black polyethylene bags and remained out of the range of sunlight, which helped ensure successful germination by six days. The seedlings were fertilized twice weekly with Poly-Feed 12-43-12 (1.5 g L^{-1}), Root-Factor® (1.0 mL L^{-1}), or Maxirad® (0.5 mL L^{-1}). Upon reaching 20 cm in height, the seedlings were transplanted into growth bags (Germinaza®, 100 cm×16 cm) filled with coconut fiber and dust (70: 30%), three plantlets were transplanted into each growth bag at 30 cm of distance between plants, and the growth bags were deposited into the gothic greenhouse. Six furrows were established with 18 growth bags and 54 *C. chinense* plants, and the distance between the furrows was 1.3 m, resulting in a planting density per hectare of 25,308. A drip irrigation system was established by installing a 1/4-inch hose, a 2 L h^{-1} dripper and 35 cm microtube for each plant. Two independent irrigation systems were installed to control the amount of water for the irrigation sheets at 75% and 100%. Each irrigation system had a 2 HP electric hydro pump (Aquapak, 2HP) adapted to a timer to automate the application of water and fertigation. The average water demand per plant ranged from 1.2 to 3.9 L day^{-1}.

Nutritional plan

A fertilization plan of 176.5–130.0–254.0 kg ha^{-1} for N, P and K was used. The nutrient sources (N-P-K) were phosphonitrate (30-3-0), potassium sulfate (0-0-51+18 S), and monopotassium phosphate (0-52-34). Turgent-Ca® (AgroScience, USA) was used for Ca contribution. Micronutrients (B, Cu, Fe, Mn, Mo, and Zn) were supplied by FullMix B® (Green How, USA). Fertilization was divided into four stages: 1) Adaptation: A fertilization rate of 2.0-1.0-1.0 (N-P-K) was applied for 21 days during six fertigations. 2) For development, a fertilization rate of 3.0-1.0-2.0 (N-P-K) was applied for 30 days in 10 fertigations. 3) Fruiting, a fertilization rate of 2.0-3.0-2.0 (N-P-K) was applied for 35 days in 12 fertigations. Finally, 4) for production, a fertilization rate of 1.0-1.0-4.0 (N-P-K) was applied for 65 days in 28 fertigations. All fertilizers were applied by fertigation.

Treatments and experimental design

The study was established through an A × B factorial experimental design, where Factor A was the irrigation level at 100% and 75%, and B was the inoculation of the EF (*B. brongniartii* and *P. lilacinum*) and a control (without application). The experiment included six treatments: T1) 100% water, T2) 100% water + *P. lilacinum*, T3) 100% water + *B. brongniartii*, T4) 75% water, T5) 75% water + *P. lilacinum*, and T6) 75% water + *B. brongniartii*. Each treatment consisted of 15 repetitions. The applications of the HFs in the treatments were carried out every seven days during the first month after transplantation. For each plant, 50 mL of a solution of conidia (active agent) was applied by drenching at a concentration of $1×10^7$ conidia mL^{-1}.

Agronomic response variables

Plant height was measured from the base of the plant to the apical branch using a flexometer (cm); stem diameter was measured using a digital Vernier (mm) at 10 cm from the soil. The chlorophyll index was evaluated with a FieldScout® CM 1000 chlorophyll meter (index). This procedure was carried out at 11:00 h in the morning at approximately 20 cm between the meter and the leaf beam, three measurements were made to obtain the mean data. The leaf area (cm^2) was measured with a portable leaf area meter (CID Bio-Science®, Washington, USA), and three leaves per plant were taken from the middle part of the plant. This variable was measured at 60 dat.

Fruit quality and yield

One hundred fruits were randomly taken from each treatment during the harvest stage (90 dat) to measure the fruit weight (FW in g) with an analytical balance, equatorial diameter (ED in mm), and horizontal diameter (HD in mm) through a digital Vernier. For yield, fruits were harvested at their point of maturity, that is, when the orange color appeared, and from 75 days onward. The number of fruits per plant was counted (fruits plant^{-1} in number) and weighed (in g plant^{-1}), and the yield per ha was calculated in each repetition by multiplying the yield per plant by the plantation density per ha (25,308 plants ha^{-1}).

Data analysis. The agronomic, fruit quality, and yield data were analyzed with a test of homogeneity of variance (P<0.05), analysis of variance (P<0.05) and multiple comparisons of means (P<0.05) using the Tukey test. All analyses were performed in Statgraphics V8 for Windows.

Results

Plant height

Plant height significantly differed at 7 (P<0.00001), 14 (P<0.0006), and 30 (P<0.0145) days after transplant (dat) for the EF factor (Table 1). Compared with noninoculated plants, both *B. brongniartii*- and *P. lilacinum*-inoculated plants were taller at 7 and 14 dat. The plants inoculated with *B. brongniartii* reached heights of 14.1 and 16.5 cm, and the plants inoculated with *P. lilacinum* reached heights of 12.8 and 15.1 cm, whereas the noninoculated plants reached heights of 9.5 and 12.5 cm, respectively. No significant differences were observed in the remaining sampling dates between 45 and 90 days after planting (P>0.05). The amount of water had no effect on plant height during the first three samplings at 7, 15, and 30 dat (P>0.05). Conversely, plants with a lower irrigation rate (75%) were shorter than plants with full irrigation (100%). The height of the plants under irrigation deficit was reduced by 15.7%, 30.0%, 27.45%, and 22.25% at 45, 60, 75, and 90 dat, respectively (Table 1).

Table 1. Plant height (cm) of habanero chili (*Capsicum chinense*) var. Chichen Itza inoculated with entomopathogenic fungi under water stress.

Factors	Days after transplant						
EF	7	14	30	45	60	75	90
B. brongniartii	14.1±0.70 a	16.5±0.71 a	30.8±1.2 a	49.6±1.6	82.8±2.9	99.2±3.2	118.3±3.3
P. lilacinum	12.8±0.65 a	15.1±0.60 a	27.7±1.1 ab	48.3±1.6	84.1±2.9	99.0±2.7	117.2±2.7
Without inoculation	9.5±0.72 b	12.5±0.68 b	26.6±1.0 b	48.3±1.5	84.9±2.7	100.0±2.8	116.8±2.6
Water stress							
100%	12.1±0.64	14.5±0.65	29.5±1.05	52.3±1.3 a	94.9±2.0 a	111.4±1.9 a	129.1±1.7 a
75%	12.2±0.51	14.8±0.54	27.3±0.86	45.2±0.9 b	73.0±1.5 b	87.4±1.5 b	105.6±5.6 b
Interactions							
B. b + 100%	12.8±1.15 abc	15.4±1.14 abc	29.4±2.0 ab	52.1±2.4 a	91.2±4.3 a	109.1±4.3 a	128.4±4.3 a
B. b +75%	15.2±0.72 a	17.5±0.81 a	31.7±1.4 a	47.2±1.8 ab	74.4±2.8 b	89.4±3.1 b	107.6±3.1 b
P. l + 100%	14.3±0.93 ab	16.2±0.89 ab	30.3±1.8 ab	53.1±2.0 a	98.8±2.1 a	112.8±2.8 a	133.4±1.9 a
P. l + 75%	11.3±0.75 bcd	14.1±0.74 abc	25.1±1.2 ab	43.5±1.6 b	69.4±2.8 b	85.2±2.4 b	101.1±2.1 b
100%	9.1±0.78 d	12.1±1.0 c	28.2±1.7 ab	51.7±2.3 ab	94.7±3.4 a	112.5±2.3 a	125.5±2.3 a
75%	10.0±0.59 cd	12.9±0.85 bc	25.0±1.2 b	44.9±1.7 ab	75.0±2.0 b	87.6±2.5 b	108.2±2.6 b
P values							
EF	0.00001	0.0002	0.0294	0.7509	0.8472	0.9603	0.9633
Water stress	0.8851	0.6768	0.951	0.00001	0.00001	0.00001	0.00001
Interactions	0.00001	0.0006	0.0145	0.0023	0.00001	0.00001	0.00001

Means with different letters are significantly different from each other, according to the Tukey comparison of means (P=0.05). W=water amount, B. b=*Beauveria brongniartii*, P. l=*Purpureocillium lilacinum*, EF=entomopathogenic fungi.

Among the interactions, significant differences were found across all the samplings conducted ($P<0.05$). In the first two samplings (7 and 15 dat), plants in treatments B. b + 75% irrigation (7 dat=15.2 cm and 15 dat=17.5 cm) and P. l + 100% irrigation (7 dat=14.3 cm and 15 dat=16.2 cm) presented greater plant heights than did those in the treatments without inoculation with 100 (7 dat=9.1 cm and 15 dat=12.1 cm) and 75% irrigation (7 dat=10.0 cm and 15 dat=12.9 cm). Moreover, at 30 dat, the B. b + 75% irrigation treatment resulted in taller plants, at 31.7 cm, whereas the noninoculated plants + 75% irrigation resulted in shorter plants (plant height of 25.0 cm). Inoculation with B. b. (47.2 cm) and P. l. (43.5 cm) in plants with 75% irrigation led to statistically similar plant heights as those of plants with 100% irrigation (51.7 cm). In the last three evaluations (60, 75, and 90 days after planting), the treatments with full irrigation at 100% allowed greater plant height than did the treatments with 75% irrigation; the inoculation of microorganisms had no effect on these evaluations (Table 1).

Stem diameter

Significant differences in plant diameter at 7 ($P<0.0190$), 14 ($P<0.0422$), 30 ($P<0.0008$), and 45 ($P<0.0034$) dat (Table 2) were observed for the EF factor. Compared with noninoculated plants, Habanero pepper plants inoculated with *B. brongniartii* presented increases in diameter at 7 (2.67 mm) and 14 dat (2.99 mm). Conversely, the evaluation at 30 and 45 dat indicated that, compared with noninoculated plants, plants inoculated with *P. lilacinum* presented 14.3% and 10.0% greater stem diameter growth, respectively, and those inoculated with *B. brongniartii* presented 9.15% greater stem diameter growth. Moreover, the last three samplings presented no significant differences among the evaluations ($P>0.05$).

The irrigation level did not lead to significant differences across the evaluations at 7 dat ($P>0.05$). In contrast, the sampling at 15 dat revealed that plants with a lower irrigation percentage (75%) presented greater plant diameter (2.95 mm) than

Table 2. Stem diameter (mm) of habanero chili plants (*Capsicum chinense*) var. Chichen Itza inoculated with entomopathogenic fungi under water stress.

Factors	Days after transplant						
EP	7	14	30	45	60	75	90
B. brongniartii	2.67±0.06 a	2.99±0.09 a	5.90±0.19 b	7.42±0.17 ab	9.78±0.23	11.33±0.15	12.24±0.17
P. lilacinum	2.59±0.05 ab	2.78±0.09 ab	5.63±0.15 b	7.18±0.13 b	9.71±0.29	11.42±0.26	12.24±0.25
Without inoculation	2.41±0.07 b	2.63±0.11 b	6.44±0.15 a	7.90±0.14 a	10.16±0.15	11.28±0.20	12.12±0.18
Water stress							
100%	2.50±0.06	2.62±0.10 b	6.09±0.15	7.40±0.12	10.33±0.16 a	11.70±0.16 a	12.73±0.16 a
75%	2.61±0.04	2.95±0.05 a	5.89±0.11	7.60±0.13	9.47±0.19 b	10.99±0.15 b	11.73±0.13 b
Interactions							
B. b + 100%	2.58±0.11	2.82±0.15 ab	6.00±0.21 ab	7.18±0.21 b	9.84±0.32 a	11.48±0.25 ab	12.75±0.25 ab
B. b +75%	2.76±0.07	3.16±0.08 a	5.79±0.22 ab	7.67±0.25 ab	9.80±0.33 a	11.17±0.17 ab	11.73±0.13 bc
P. l + 100%	2.52±0.08	2.66±0.16 ab	5.74±0.25 ab	7.30±0.23 ab	10.87±0.27 a	12.20±0.27 a	13.02±0.28 a
P. l + 75%	2.66±0.06	2.90±0.09 ab	5.53±0.18 b	7.06±0.14 b	8.55±0.28 b	10.64±0.35 b	11.45±0.32 c
100%	2.41±0.13	2.46±0.19 b	6.52±0.31 a	7.71±0.18 ab	10.28±0.20 a	11.41±0.31 ab	12.40±0.31 abc
75%	2.40±0.08	2.80±0.10 ab	6.37±0.18 ab	8.08±0.21 a	10.04±0.24 a	11.16±0.25 ab	11.84±0.19 bc
P values							
EP	0.0190	0.0422	0.0008	0.0034	0.2502	0.8756	0.8672
Water stress	0.1894	0.0822	0.2633	0.2288	0.0003	0.0023	0.00001
Interactions	0.0563	0.0212	0.0076	0.0070	0.00001	0.0067	0.0001

Means with different letters are significantly different from each other, according to the Tukey comparison of means (P=0.05). B. b=*Beauveria brongniartii*, P. l=*Purpureocillium lilacinum*, EF=entomopathogenic fungi.

did plants with full irrigation (100%) (2.62 mm). At 30 and 45 dat, the two irrigation levels evaluated exhibited no significant differences. The stem diameter values of the plants subjected to irrigation deficit were 9.1%, 8.43%, and 8.5% smaller at 60, 75, and 90 dat, respectively (Table 2).

No significant differences were observed in the interactions between the first sampling (7 dat) and the remaining evaluations (P>0.05). At 14 days after transplanting, the B. b + 75% irrigation treatment had a significantly greater effect on stem diameter (3.16 mm) than the treatments without microorganisms or full irrigation (100% and 2.6 mm, respectively). Furthermore, the data obtained on Day 30 after transplanting revealed that, compared with those in the noninoculated treatment + full irrigation (100%), the diameter of the plants in the P. l + 75% irrigation treatment was 17.9% lower. Compared with the B. b + 100% irrigation (7.18 mm) and P. l + 75% irrigation (7.06 mm) treatments, the noninoculated treatment + 75% irrigation yielded a greater stem diameter (8.08 mm) in the sampling conducted 45 days after transplanting. Compared with the other treatments at 60 dat, the P. l + 75% irrigation treatment resulted in the smallest stem diameter of 8.55 mm compared to the values of 9.80, 9.84, 10.04, 10.28, and 10.87 mm. Finally, the P. l + 100% treatment led to an increase in stem diameter at 75 (12.20 mm) and 90 (13.02 mm) dat compared to the P. l + 75% irrigation treatment, in which the stem diameter was 14.6% and 13.71% lower, respectively (Table 2).

Chlorophyll index (CI)

B. brongniartii and *P. lilacinum* treatment led to no significant differences in CI (P>0.05); both led to the synthesis of the same amount of pigment as in noninoculated plants at 7, 14, and 30 dat (Table 3). Compared with that of noninoculated plants, the chlorophyll index of habanero pepper plants was significantly higher (P=289.1) with *B. brongniartii* inoculation (P=248.8) 45 days after transplanting. However, the evaluation at 60 dat revealed no significant differences. Sampling at 75 dat revealed a greater CI (348.5, 339.3) in plants inoculated with both fungal species than in noninoculated plants.

Table 3. Chlorophyll index in habanero pepper plants (*Capsicum chinense*) var. Chichen Itza inoculated with entomopathogenic fungi and subjected to irrigation deficit.

Factors	Days after transplant						
EF	7	14	30	45	60	75	90
B. brongniartii	123.5±3.4	156.2±6.0	298.9±9.3	289.1±12.3 a	357.9±13.4	348.5±14.0 a	354.4±10.5
P. lilacinum	116.7±3.2	153.5±6.5	277.9±11.2	271.9±12.5 ab	325.0±19.0	339.3±16.8 a	317.4±9.7
Without inoculation	117.8±2.4	148.4±5.0	289.3±18.9	248.8±9.2 b	313.6±14.2	296.5±20.7 b	332.2±10.6
Water stress							
100%	122.1±2.5	157.9±3.7	302.2±12.0	275.6±9.4	338.0±12.9	375.7±13.5 a	331.7±9.8
75%	116.6±2.3	147.4±5.6	275.1±10.0	264.2±9.8	326.3±13.2	280.4±12.7 b	343.4±7.3
Interactions							
B. b + 100%	129.1±3.3	154.9±4.9	283.4±14.0 ab	270.0±18.5 ab	340.0±17.1	353.6±22.3 ab	358.13±19.0
B. b +75%	117.9±5.8	157.4±11.2	314.4±11.3 ab	308.2±15.3 a	375.7±20.2	343.4±17.7 ab	350.6±8.6
P. l + 100%	120.4±5.6	160.0±6.0	284.5±15.7 ab	290.6±16.6 ab	356.4±31.5	397.0±20.8 a	300.8±9.8
P. l + 75%	117.9±2.8	147.0±11.7	271.4±16.6 ab	253.1±18.1 ab	293.6±21.4	281.6±21.5 b	334.0±12.8
100%	116.3±3.7	159.0±8.3	338.8±28.0 a	266.2±13.6 ab	317.5±15.9	376.6±27.0 ab	336.2±15.5
75%	118.9±3.2	137.8±4.6	239.6±18.5 b	231.4±11.2 b	309.7±24.5	216.4±11.9 c	345.6±15.1
P values							
EF	0.2430	0.6394	0.5167	0.0428	0.1629	0.0128	0.1060
Water stress	0.1246	0.1257	0.0708	0.3805	0.5519	0.00001	0.4174
Interactions	0.1769	0.3800	0.0054	0.0170	0.1513	0.00001	0.2590

Means with different letters are significantly different from each other, according to the Tukey comparison of means (P=0.05). B. b=*Beauveria brongniartii*, P. l=*Purpureocillium lilacinum*, EF=entomopathogenic fungi.

However, the last sampling at 90 dat indicated that the CI of the habanero pepper plants did not significantly differ (P>0.05) (Table 3).

The amount of irrigation applied to the habanero pepper plants did not influence the CI; therefore, the CI did not significantly differ at 7, 14, 30, 45, and 60 days after transplanting (P>0.05). The values for full irrigation (100%) were 122.1, 157.9, 302.2, 275.6, and 338.0 units, whereas the values for plants with irrigation deficit (75%) were 116.6, 147.4, 275.1, 264.2, and 326.3 units. However, the sampling on Day 75 after transplanting reflected a greater CI in plants with full irrigation (100%, Table 3).

The CI variable showed no significant differences at 7 and 14 days after transplanting in any of the interactions. However, at 30 dat, the treatment without inoculation or full irrigation (100%) led to a greater CI (338.8 units) than did the treatment without microorganisms or irrigation deficit (75%, 239.6 units). Nevertheless, the evaluation on Day 45 after transplanting indicated that, compared with B. b +75% irrigation (308.2 units), the treatment without inoculation + 75% irrigation (231.4 units) considerably reduced CI levels by 33.1%. Sampling at 65 dat revealed that none of the treatments resulted in significant differences (P>0.05). At 75 dat, the P. l + 100% irrigation treatment (397.0 units) provided a greater CI for habanero pepper plants than did the P. l + 75% irrigation treatment (281.6 units) and the treatment without microorganisms + 75% irrigation (216.4 units, Table 3).

Fruit quality

Fruit width significantly differed (P<0.0430) with respect to the EF factor. Plants inoculated with *B. brongniartii* (26.51 mm) were statistically equal to uninoculated plants (26.41 mm), but *P. lilacinum* led to reduced FWi (25.47 mm) (Table 4). The FL and FW did not significantly differ. Full irrigation (100%) resulted in greater fruit length at 42.73 mm and a greater weight of 11.13 g. Plants with irrigation deficit presented less growth (4.0% fruit length (mm) and 5.19% weight (g)). In terms of diameter, no significant differences (P>0.05) were observed in any of the plants inoculated with microorganisms (Table 4). Fruits obtained

Table 4. Fruit quality and yield of habanero chili plants (*Capsicum chinense*) var. Chichen Itza inoculated with entomopathogenic fungi under irrigation deficit.

Factors	Fruit quality			Fruit yield		
EF	Length (mm)	Width (mm)	Weight (g)	Fruits plant^{-1}	g plant^{-1}	t ha^{-1}
B. brongniartii	42.3±0.5	26.5±0.3 a	10.9±0.1	37.2±2.0	397.0±20.8	8.8±0.5
P. lilacinum	40.9±0.5	25.5±0.3 b	10.6±0.2	39.7±3.1	387.7±27.0	8.6±0.6
Without inoculation	42.5±0.4	26.7±0.4 a	11.0±0.1	40.9±1.8	436.2±20.3	9.7±0.4
Water stress						
100%	42.7±0.4 a	26.5±0.3	11.1±0.1 a	40.68±2.0	416.8±19.2	9.3±0.4
75%	41.1±0.4 b	25.9±0.3	10.6±0.1 b	37.89±1.9	397.2±18.5	8.9±0.4
Interactions						
B. b + 100%	43.0±0.7 a	26.7±0.5	11.1±0.2 ab	34.66±3.0	369.6±31.0	8.2±0.7
B. b +75%	41.6±0.7 ab	26.2±0.4	10.8±0.2 ab	39.73±2.7	424.4±27.0	9.5±0.6
P. l + 100%	42.3±0.6 ab	25.6±0.5	10.9±0.3 ab	42.8±4.1	394.0±32.3	8.8±0.7
P. l + 75%	39.5±0.7 b	25.3±0.4	10.3±0.2 b	36.6±4.5	381.5±44.5	8.5±0.9
100%	42.8±0.7 a	27.1±0.5	11.4±0.2 a	44.6±2.9	486.7±30.1	10.8±0.6
75%	42.1±0.6 ab	26.1±0.5	10.6±0.2 b	37.33±2.2	385.6±20.9	8.6±0.4
P values						
EP	0.0580	0.0430	0.1686	0.5228	0.2759	0.2759
Water stress	0.0048	0.1274	0.0019	0.3093	0.4518	0.4518
Interactions	0.0078	0.1076	0.0084	0.2711	0.1123	0.1123

Means with different letters are significantly different from each other, according to the Tukey comparison of means (P=0.05). B. b=*Beauveria brongniartii*, P. l=*Purpureocillium lilacinum*, EF=entomopathogenic fungi.

from habanero pepper plants with the interaction of the treatment without microorganisms + 100% irrigation (42.85 mm) and B. b + 100% irrigation (43.01 mm) exhibited greater fruit length than the fruits from P. l + 75% irrigation (39.5 mm). Plants inoculated with *B. brongniartii* with 75% (10.85 g) irrigation yielded the same FW as that of the plants fertigated at 100% (11.45 g); in this sense, inoculation with *B. brongniartii* allows the relief of plant stress despite water deficit (Table 4).

Fruit yield

The fruit yield of the habanero pepper plants was not significantly different ($P>0.05$) for any of the variables (fruits plant^{-1}, g plant^{-1}, t ha^{-1}); the number of fruits ranged from 37.20 to 39.70 for the plants inoculated with *B. brongniartii* and *P. lilacinum*, respectively. Moreover, the number of fruits in the noninoculated plants was 40.96 (Table 4). The irrigation regimens did not significantly affect fruit yield; the plants produced between 40.68 and 37.80 fruits plant^{-1}, 397.15 and 416.77 g plant^{-1}, and 8.65 to 9.73 t ha^{-1} (Table 5). Finally, the interaction effect between the EF and irrigation regimens did not significantly differ ($P>0.05$, Table 4).

Discussion

Beauveria brongniartii had favorable effects on the vegetative growth and yield of *Capsicum chinense* var. Chichen Itza under water deficit and greenhouse conditions. The habanero pepper plants presented a significant increase in growth at 30 dat, which, according to Toscano-Verduzco et al. (2019), may be attributed to the production of phytohormones, such as indole-3-acetic acid, that promote plant development and growth. According to Barra-Bucarei (2020), the application of EFs to plants activates mechanisms that may allow greater expression of plant growth potential and increased tolerance to abiotic stresses such as drought. The use of *B. brongniartii* as a plant growth promoter has rarely been studied; however, a study in *Vicia faba* L. reported a significant increase in fresh shoot weight when inoculated with *B. brongniartii* (Jaber & Enkerli, 2016). In *C. papaya,* inoculation with *B. brongniartii* significantly increased seed germination, plantlet height, and fresh shoot and root weights (Barajas-Méndez et al., 2022).

EFs have not been studied in depth under water deficit conditions. However, under reduced fertilization (75%), the inoculation of *B. brongniartii* in *C. chinense* plants did not increase the agronomic parameters, and at 100% fertilization, the inoculation of *B. brongniartii* increased the foliar N content, plant height, FW, FL and FWi (Toscano-Verduzco et al., 2019). Tall and Meyling (2018) reported a similar result with *Beauveria bassiana* Bals. Vuill. as a seed treatment for promoting the growth of *Z. mays*; when nutrients are abundantly available, the fungus provides growth benefits to the maize plants.

In the first three samplings, *P. lilacinum* and *B. brongniartii* were able to increase the stem diameter of the habanero pepper plants. Both *P. lilacinum* (Moreno-Salazar et al., 2020) and *B. brongniartii* (Toscano-Verduzco et al., 2019) have been characterized as phosphorus solubilizers, and both fungi are able to obtain PO_4^{3-} from $Ca_3(PO_4)_2$ and $FePO_4$ by reducing the soil pH through acid organic and siderophore production. This ability of EFs allows them to promote or maintain the growth of *C. chinense* plants, even under water deficit conditions.

In addition to increasing the vegetative growth of habanero pepper plants, *P. lilacinum* and *B. brongniartii* mitigated the water stress observed during the samplings, equating it to full irrigation. As mentioned by Raya-Díaz (2017), both entomopathogenic fungi can protect plants against abiotic stresses and improve their ecological adaptability, increasing their tolerance to environmental stresses such as drought, temperature, and nutritional stress. Thus, *Beauveria* entomopathogenic fungi promote growth, improving the vigor of cabbage plants under water stress (Dara et al., 2017). Recently, Gana et al. (2022) assessed the effects of water deficiency (watering intervals) and the inoculation of *B. bassiana* on the plant growth, nutrient uptake, secondary metabolite contents, and antioxidant capacity of *Allium cepa* L. Their results indicated that plants inoculated with *B. bassiana* under water stress showed an increase of P, K, and Fe uptake; polyphenol activity was significantly greater in the plants treated with *B. bassiana* than in the non-fungus-treated plants. In addition, water stress in inoculated plants positively influenced the flavonol content in onion bulbs (Gana et al., 2022). In this study, plant height and stem diameter were positively affected by *B. brongniartii* in the first stage of plant growth (0-30 dat).

In another study, Jaramillo-Salazar (2018) reported that plants respond morphologically and physiologically to the environmental conditions in which they develop, thus modifying the amount of pigments. Plants subjected to any type of stress tend to lose their photosynthetic capacity, and the chlorophyll content in their leaves decreases. In terms of the chlorophyll content at 45 and 75 dat, *P. lilacinum* had positive effects on increasing the chlorophyll content. Similar results were obtained by Chiquito-Contreras (2018), who reported that the chlorophyll content increases in plants due to inoculation with arbuscular mycorrhizal fungi (AMF) and bacteria, increasing their photosynthetic capacity.

Water deficit in *C. chinense* decreased the FL and FW; however, the interaction between *B. brongniartii* and 75% irrigation resulted in the same FW as irrigation at 100%. The hyphae and mycelia of soil-borne micromycetes, including EFs, can transport nutrients for plants (Dellagi et al., 2020), especially when they are endophytes, such as those in the genus *Beauveria*, which may constitute a mechanism to improve or maintain the physiological quality of plants under drought conditions. In this sense, *B. bassiana* can improve the chlorophyll index (SPAD units) in barley varieties when inoculated (1×10^7 conidia mL^{-1}) and establish itself in an endophytic manner (Veloz-Badillo et al., 2019). Finally, EFs did not improve the fruit quality or yield of *C. chinense*, contrary to Cordeiro et al. (2019), who reported that the application of arbuscular mycorrhizal fungi provides multiple benefits to the quality of strawberry fruits, including increases in pH, soluble solids content, titratable acidity and phenolic compound content. These findings also suggest that AMF influence fruit filling and the distribution of photosynthates along with improving the acquisition of nutrients and water.

In conclusion, inoculation with *B. brongniartii* improved the height and stem diameter of *C. chinense* plants in the adaptation stage. A 25% reduction in irrigation negatively affected the height and stem diameter of *C. chinense* plants. *B. brongniartii* increased the chlorophyll index in inoculated plants; in contrast, the reduction in irrigation negatively affected the chlorophyll index. The fruit quality and yield of *C. chinense* did not increase because of inoculation with entomopathogenic fungi.

Competing interest statement

The authors declare that there are no competing interests regarding the research reported in this manuscript.

Data availability statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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