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Research Article

Morphological characterization and biostimulation of growth, production, and quality of 'chiltepín' (*Capsicum annuum* var. *glabriusculum*) using an agroecological approach

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Abstract

Of the primary challenge faced in the domestication of chiltepín (*Capsicum annuum* L. var. *glabriusculum* [Dunal] Heiser & Pickersgill), the specific requirements of its natural habitat are most important. Therefore, the objective of this research was to conduct a morphological characterization of chiltepín and evaluate the impact of biostimulants on its growth, production, and quality when cultivated outside its native environment. The study was conducted using a completely randomized block design with four treatments (T1-BF, T2-RT, T3-RB, and T4-Ctrl), four replications, and five plants per experimental unit. Morphological analyses revealed significant phenotypic variability between the wild and domesticated varieties, reflecting ecological adaptations and selective pressures that have shaped its evolution. Additionally, the chiltepín showed an outstanding response to biostimulant treatments, particularly T1-BF and T2-RT; these treatments had a significant impact on plant growth and development, improving parameters such as height, stem diameter, chlorophyll content, and canopy density during the first 30 days after transplanting (DAT). Furthermore, these treatments enhanced fruit production and quality, increasing fresh weight (47%), dry weight (46%), length, and length-to-width ratio. These results not only highlight the value of the knowledge gained about the phenotypic features of chiltepín, but also the potential of the agroecological approach using biostimulants to optimize the development, production, and quality of this crop.

Keywords: agronomic parameters; biostimulants; chiltepín domestication; fruit quality; production; wild chili pepper

Introduction

Chiltepín (*Capsicum annuum* L. var. *glabriusculum* [Dunal] Heiser & Pickersgill), belonging to the Solanaceae family, has been recognized as the wild ancestor of globally cultivated chili pepper species (Mares-

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Quiñones and Valiente-Banuet, 2019). This species is highly valued for its popularity as a condiment in Mexican cuisine, attributed to its distinctive flavor and high capsaicin content, averaging 150,000 Scoville heat units (Hayano-Kanashiro *et al.*, 2016; Hernández-Pérez *et al.*, 2020).

Chiltepín is widely distributed across coastal and mountainous regions of Mexico, ranging from the Pacific Ocean to the Gulf of Mexico, and extending from Sonora to Chiapas and from Tamaulipas to the Yucatán Peninsula (Luna *et al.*, 2018; González-Jara *et al.*, 2011). In the state of Chihuahua, it thrives in arid and semi-arid areas, primarily within xerophilous scrublands and oak forests located in the municipalities of Batopilas, Chínipas, and Urique, at altitudes ranging from 400 to 2500 meters above sea level (INEGI, 2010).

Chiltepín is one of the most challenging crops to domesticate due to its strict edaphoclimatic requirements, among others (Beltrán-Burboa *et al.*, 2020). In its natural environment, chiltepín thrives in soils rich in organic matter and under the shade of trees, which protects the plants from adverse biotic and abiotic factors (Mares-Quiñones and Valiente-Banuet, 2019). This preference for such natural conditions has led people to value wild chiltepín more, reducing interest in its domestication (Ramírez *et al.*, 2018; Villalón-Mendoza *et al.*, 2023). The preference for wild chiltepín exerts strong pressure on the species, putting its survival at risk.

Preserving chiltepín and developing sustainable management practices are essential for successfully adapting this wild crop outside its natural habitat (Alonso *et al.*, 2008; Vera-Sánchez *et al.*, 2016). Various efforts have attempted to replicate agroforestry conditions in controlled systems, including shaded cultivation (Molina *et al.*, 2009), intensive production systems with shade nets (Mares-Quiñones and Valiente-Banuet, 2019), greenhouses (Araiza *et al.*, 2011), and mesquite stand plantations (McCaughey-Espinoza *et al.*, 2020). However, the domestication of chiltepín remains a significant challenge. In this context, biostimulants have emerged as an innovative strategy in agriculture, as they stimulate physiological processes in plants, improving nutrient absorption, resistance to biotic and abiotic stress, and overall plant productivity (du Jardin, 2015; Valverde-Lucio *et al.*, 2020). Due to the phytogenetic and economic relevance of chiltepín, the application of biostimulants in production systems outside its natural habitat could be a promising alternative for implementing sustainable and high-production agricultural practices. Therefore, the objective of this study was to determine the morphological variability and evaluate the effect of biostimulants on the development, production, and fruit quality of chiltepín cultivated using an agroecological approach.

Materials and Methods

Collection of plant material

Fruits of domesticated chili varieties and the wild chili variety were obtained from the municipalities of the State of Chihuahua, Mexico (Table 1).

Table 1. Geographical and climatic characteristics of chili pepper varieties collected from different municipalities in the state of Chihuahua, in northern Mexico

Samples	Municipalities	GL	MASL	CT	AAP (mm)	AAT (°C)
Chiltepín (CHB)	Batopilas	27°01'50" N and 107°43'56" W	636	Semi-warm	850	23.8
Árbol (A)	Aldama	28°35′40.92″N, 105°34′15.6″W	1,119	Desert	318	19.5
Negro (N)	Julimes	28°32′0″N, 105°3′0″W	1,700	Hyper-arid	60	18.3
Mirasol (M) & Güerito (G)	Delicias	28°11′36″N, 105°28′16″W	1,170	Semi-arid	334	18.8

GL = geographic location, MASL = meters above sea level, CT = climate type, AAP = average annual precipitation, AAT = average annual temperature.

In each municipality, three samples of fresh red fruits (1 kg each) were collected in pre-labeled plastic bags. The samples were transported to the Laboratory of Applied Microbiology, Phytopathology, and Postharvest Physiology (MAFFP) at the Department of Agrotechnological Sciences of the Autonomous University of Chihuahua. Fruits were left on a laboratory bench at 24 ± 2 °C until dried. Dry fruits were crushed, and healthy and uniformly size seeds were selected and stored at 4 °C for further analysis.

Seedling production

Following a viability test using the flotation method (Beltrán-Barboa *et al.*, 2020), viable chiltepín seeds were disinfected in a 7% sodium hypochlorite (Cloralex®) solution with continuous agitation for 10 minutes. The solution was then discarded, and the seeds were rinsed three times with sterile distilled water for 10 min each. The disinfected seeds were then incubated in an acidic solution under static and dark conditions at 24 ± 1 °C for 24 hours. After incubation, the solution was discarded, and the seeds were dried on sterile blotting paper. Ten sterile petri dishes, each lined with a sterile filter paper disk, were prepared, and 20 seeds were placed in each dish. The filter paper was moistened with sterile distilled water, and the petri dishes were sealed with adherent film. They were then transferred to a plant germination chamber set to a photoperiod of 16 hours light at 25 ± 1 °C and 8 hours darkness at 18 ± 1 °C for 20 days.

The *in vitro* germinated chiltepín seeds were transferred to germination trays with 98 cavities filled with a Sunshine Mix 3 peat moss and compost in a 3:1 ratio. The trays were placed in a germination chamber at 24 ± 1 °C with a photoperiod of 16 h light and 8 h darkness. Watering was performed with filtered water every 3 days during the initial weeks and then every 8 days until the plants developed 10 true leaves. The seed disinfection process for domestic chili varieties was the same as that described for wild chili seeds. After disinfection the seeds were sown in 98-cell trays filled with a Sunshine Mix 3 peat moss substrate. Two disinfected seeds were placed in each cavity at a depth of 0.5 cm and watered to field capacity. The trays were then placed in the plant germination chamber. Watering was performed every 8 days until the seedlings developed 10 true leaves. Seedlings of both domestic and wild varieties were transferred to small plastic pots (12 x 12 x 12 cm) filled with a 1:1 ratio of soil and Sunshine Mix 3 peat moss substrate.

The pots were placed in a greenhouse at 28 ± 2 °C, where watering and fertilization were carried out simultaneously every three days for 12 weeks. The nutrient solution was formulated in the MAFFP laboratory and contained 1.5 $g{\cdot}L^{-1}$ of 12-61-00 (N-P-K), 1.5 $mL{\cdot}L^{-1}$ of Ca, and 1.5 $g{\cdot}L^{-1}$ of 18-18-18 (N-P-K). Additionally, 3 $mL{\cdot}L^{-1}$ of Nutrisorb®, 2 $mL{\cdot}L^{-1}$ of Radigrow®, and 3 $mL{\cdot}L^{-1}$ of ATPUP® were added during irrigation.

Morphological variability

The morphological variability of wild chiltepín was assessed following the methodology proposed by the International Plant Genetic Resources Institute (IPGRI, 1995). Characterization was carried out during the seedling, plant, inflorescence, and seed stages, considering the primary qualitative traits used to describe *Capsicum* species. To assess morphological variability, domesticated chili varieties (Árbol, Mirasol, Güerito, and Negro) were used for comparison. The descriptors included cotyledon leaf colour, node anthocyanin, stem pubescence, growth habit, leaf colour, leaf shape, fruit shape, fruit apex shape, and seed colour. Statistical data analysis was conducted using RStudio, version 1.2.5033 (R Core Team, 2024). This analysis examined and visualized associations between categories, and Chi-square tests were performed to determine statistical differences ($\alpha = 0.05$).

Evaluation of biostimulants on the growth and production of chiltepín

Location and orchard preparation

The study was conducted from June to November 2020 in an orchard located at the facilities of the Department of Agrotechnological Sciences of the Autonomous University of Chihuahua. The site is situated at coordinates 28° 39’ 25” N and 106° 05 ’14” W, at an altitude of 1,440 meters above sea level, in a semi-arid climate with average annual precipitation of 400 mm and an average annual temperature of 18.2 °C (INEGI, 2010). The orchard was established over an area of 121 m^2. The site was cleared, and an 8 × 8 m (64 m^2) quadrant was delineated, leaving a 1.5 m buffer on each side of the original space. Within this inner quadrant, 16 blocks, each measuring 1.55 m^2, were arranged with 0.6 m spacing between them. Each block underwent double digging following the methodology proposed by Jeavons (2001). The topsoil layer was removed to a depth of 30 cm using a square shovel, while the second layer was further loosened to an additional depth of 30 cm with a digging fork. After double digging, the soil was sieved through a metal mesh with a 1 cm^2 grid to create a more uniform and porous substrate. A drip irrigation system was installed, with emitters delivering a flow rate of 4 $L{\cdot}h^{-1}$, positioned at each plant's location. Five well-spaced plants were transplanted into each block. To protect the orchard from animal intrusion and potential damage, it was enclosed within a wire mesh fence.

Experimental design

To assess the effect of biostimulants on the growth, development, production, and quality of chiltepín under open-field conditions, a completely randomized block design was used. The experiment included five treatments (Table 2) and four replications, with each replication consisting of five plants as the experimental unit.

Statistical data analysis was conducted using the InfoStat software package (Di Rienzo *et al.*, 2011). An analysis of variance (ANOVA) was performed, followed by mean comparison analysis using Fisher's LSD test at a significance level of $\alpha = 0.05$, n = 4.

Table 2. Definition of treatments based on different types of biostimulants under the spring-winter cycle

Treatments	**Biostimulants**		
	Active Ingredient (IA)	**Commercial name**	**Dose ($g{\cdot}L^{-1}$)**
T1-BF	Exuroot, *Trichoderma harzianum, Bacillus subtilis, Paecilomyces* sp.*, Penicillium* sp.*, Azospirillum* sp. and *Azotobacter* sp.	Biofit® RTU	8.0
T2-RT	Exuroot, *T. harzianum* and *B. subtilis*	Rhizo TX®	8.0
T3-RB	Exuroot and *Bacillus cereus* var. *mycoides*, *B. megaterium* and *B. subtilis*	Rhizobac® Combi	8.0
T4-Ctrl	Without biostimulants	Control	0.0

Experiment establishment and treatment application

The experiment was initiated in June to prevent damage from late frosts. In each block, five seedlings with ten true leaves were transplanted. Treatments were applied to the soil around the base of the seedling stems. The first application was carried out at the time of transplanting (June 15), followed by four additional applications at 15-day intervals. Given the low precipitation levels in the state of Chihuahua (northern Mexico), irrigation was applied for two hours every 48 or 72 h, depending on prevailing temperatures, throughout the experiment.

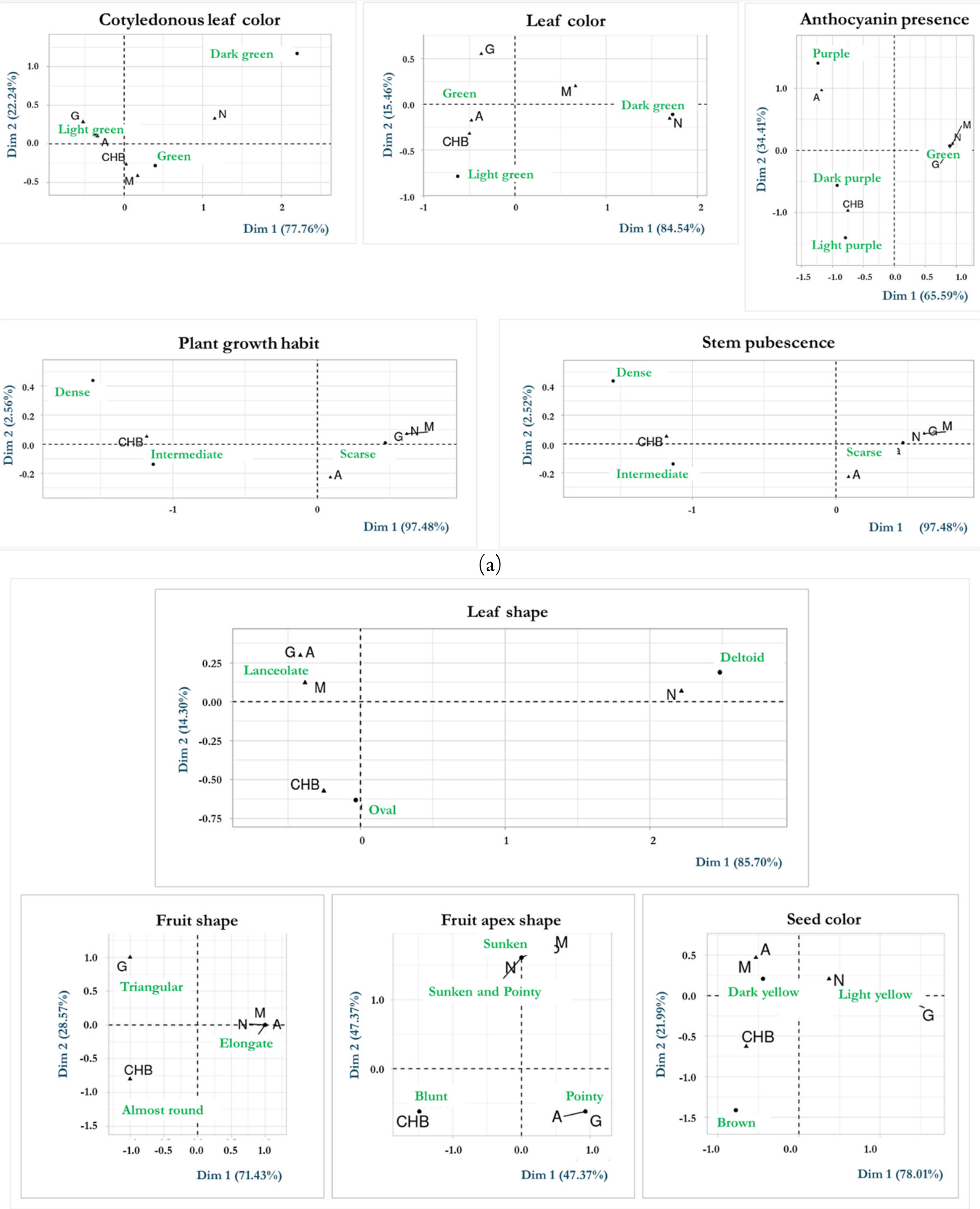
Evaluated variables

The evaluated variables included plant height (cm), stem diameter (mm), plant canopy width (cm), and chlorophyll content, measured using a chlorophyll meter (Minolta SPAD 502 Plus). Additionally, fruit-related variables were assessed, including the number of fruits, fresh fruit weight (g), dry fruit weight (g), fruit length (mm), fruit width (mm), and the fruit length-to-width ratio, calculated by dividing fruit length by fruit width. Agronomic variables were measured four times at 15-day intervals, starting 15 days after the first treatment application. Production and quality variables were assessed at the end of the harvest, based on cumulative data from three harvests.

Results and Discussion

Qualitative morphological descriptors

Analyses of the qualitative morphological descriptors in the chiltepín variety compared to domesticated chili varieties revealed significant differences among the evaluated traits (Figures 1a and 1b). These findings underscore the importance of specific traits as phenotypic markers in studies on genetic diversity, adaptability, and plant breeding.



(a)

(b)

Figure 1a, b. Comparison of qualitative traits between chiltepín and domesticated chili varieties CHB = Chiltepín, A = Árbol, G = Güerito, M = Mirasol, N = Negro

In Figure 1a, cotyledon leaf color (p = 0,1223) showed no significant differences among the analyzed varieties. This trait was homogeneously distributed, suggesting it is not a relevant discriminative marker in the populations analyzed. According to Paran and van der Knaap (2007), homogeneity in such traits may reflect conservative selection processes. Analysis of nodal anthocyanin presence revealed a highly significant value (p < 0,001), indicating heterogeneity among the varieties, suggesting that this compound may be related to adaptations to environmental conditions, given its protective role against oxidative stress (Lightbourn *et al.*, 2008; Bobadilla-Larios *et al.*, 2017). Stem pubescence also showed significant differences (p < 0,05), which suggests adaptive advantages by acting as a physical barrier against pests, a relevant mechanism in the plant's passive defense strategies (Bobadilla-Larios *et al.*, 2017). Growth habit displayed significant variability (p < 0.05) among the analyzed varieties, possibly influenced by local agroecological conditions. Indeterminate growth, typical of wild species, is associated with survival strategies in adverse environments (Sánchez and Gutiérrez, 2016; Carrillo-Montoya and Vargas-Rojas, 2023). Leaf color analysis also showed significant differences (p < 0.05), suggesting optical or photosynthetic adaptations linked to ultraviolet radiation in mountainous environments, such as those present in the regions of origin of these varieties (Gepts, 2004).

In Figure 1b, leaf shape exhibited highly significant heterogeneity (p < 0,001), underscoring its importance in adaptation to specific microclimates, particularly for wild varieties. This trait is crucial for photosynthetic efficiency (Nicotra *et al.*, 2011). Additionally, fruit shape showed high statistical significance (p < 0,001), reflecting the influence of both human and natural selective pressures. This characteristic is fundamental in genetic improvement and has significant commercial relevance due to market preferences (Paran and van der Knaap, 2007; Carrillo-Montoya and Vargas-Rojas, 2023). Fruit apex shape also demonstrated highly significant differences (p < 0,001), presenting it as a useful phenotypic marker for genetic diversity studies. This trait is associated with dehydration resistance and could provide insights into population differentiation and the evolutionary history of these varieties (Kantar *et al.*, 2017). Finally, seed color exhibited highly significant heterogeneity (p < 0,001), suggesting a possible relationship with genetic adaptations or mechanisms related to dispersal and germination, as reported in other studies (Gepts, 2004).

These results demonstrate notable morphological variability between chiltepín and domesticated chili varieties (Figures 2 and 3), underscoring the relevance of qualitative descriptors as key tools for genetic identification and characterization. This phenotypic diversity provides valuable information for genetic improvement programs, and also contributes to understanding the ecological adaptations and selective pressures that have shaped the evolution and differentiation of these varieties, particularly in response to the agroecological conditions of their environment (Cortés and López-Hernández, 2021; Bajpai *et al.*, 2022; Carrillo-Montoya and Vargas-Rojas, 2023).

Figure 2. Morphology of the wild chili variety from Batopilas, Chihuahua, Mexico; (**a**) Plantlet; (**b**) Plant in flowering and fruiting; (**c**) Plant with some ripe fruits; (**d**) Ripe fruit; (**e**) Seed (prepared by the authors)

Figure 3. Morphology of domesticated and wild chili plants, including leaves, stems, flowers, and fruits across different varieties from the state of Chihuahua (northern Mexico); (**a**) Alcala; (**b**) Árbol; (**c**) Negro; (**d**) Güerito; (**e**) Mirasol; (**f**) Chiltepín (prepared by the authors)

Effect of biostimulants on agronomic parameters of chiltepín

Biostimulation of the agronomic parameters for chiltepín during its development was most evident within the first 30 days after transplanting (DAT). At 15 DAT, significant differences ($p < 0.05$) were observed in stem diameter and plant canopy, and for both of these variables, treatment T1-BF showed superior performance compared to the other treatments. In the case of stem diameter, treatment T2-RT ranked second in efficacy, whereas for plant canopy, treatments T2-RT and T3-BR performed statistically similar to T1-BF. At 30 DAT, the effect of biostimulants was evident across all evaluated variables. Once again, treatment T1-BF statistically outperformed ($p < 0.05$) the other treatments, followed by T2-RT, which exhibited similar values to T1-BF. The control treatment consistently showed the lowest values across all evaluated variables (Table 3).

These results align with those reported by Aguirre-Medina and Espinosa (2016), particularly in the parameter of plant height, where *Azospirillum brasilense* inoculated plants reached a height of 63 cm.

The biostimulant effect on chiltepín development at 45 and 60 DAT showed no significant differences ($p > 0.05$) among treatments for any of the evaluated variables (Table 4). This pattern is common during the crop's vegetative stage. In a similar study, Sánchez and Gutiérrez (2016) reported that the effect of microorganisms such as *Azospirillum brasilense* and *Pseudomonas fluorescens* was most pronounced at 28 DAT. They also observed that growth variation was similar across treatments at 56 DAT.

Table 3. Biostimulation of chiltepín plant development at 15 and 30 days after transplanting (DAT) under open field conditions using an agroecological approach

Treatments	Variables			
	Plant height (cm)	Stem diameter (mm)	Chlorophyll content (SPAD)	Plant canopy (cm)
	Measurement at 15 DAT			
T1-BF	22.9 a	4.0 a	52.5 a	12.7 a
T2-RT	21.1 a	3.6 ab	48.3 a	10.4 ab
T3-RB	23.8 a	3.5 b	51.5 a	10.9 ab
T4-Ctrl	21.6 a	3.7 b	51.1 a	12.6 a
LSD	2.739	0.396	6.457	2.180
	Measurement at 30 DAT			
T1-BF	32.1 a	5.6 a	55.4 a	24.7 a
T2-RT	30.6 ab	5.4 ab	50.9 ab	23.5 ab
T3-RB	30.3 ab	4.9 b	48.6 b	22.5 b
T4-Ctrl	27.9 b	4.7 b	47.0 b	20.5 b
LSD	3.957	0.941	7.190	4.119

Means with a different letter(s) in each column are significantly different (LSD test, $p < 0.05$)

The vegetative phase of the crop is primarily influenced by the variety, climatic conditions, and soil properties. The variety determines the potential for development and the duration of this stage, while genetic characteristics regulate factors such as growth rate, plant architecture, and adaptability to different environments (Sánchez and Gutiérrez, 2016). In general, this phase lasts approximately 60 DAT (Álvarez and Pino, 2018), which aligns with the results of our study (Figure 3). Additionally, plant growth is influenced by climatic factors such as temperature, precipitation, and solar radiation, as well as soil characteristics, including pH, moisture, and nutrient availability (Sánchez and Gutiérrez, 2016).

Table 4. Biostimulation of chiltepín plant development at 45 and 60 days after transplanting (DAT) in open field conditions using an agroecological approach

Treatments	Variables			
	Plant height (cm)	Stem diameter (mm)	Chlorophyll content (SPAD)	Plant canopy (cm)
	Measurement at 15 DAT			
T1-BF	48.3 a	7.9 a	51.8 a	36.0 a
T2-RT	47.6 a	8.0 a	51.8 a	35.9 a
T3-RB	43.6 a	7.5 a	49.5 a	36.0 a
T4-Ctrl	43.4 a	7.0 a	48.4 a	34.1 a
LSD	6.316	1.270	4.084	8.871
	Measurement at 60 DAT			
T1-BF	72.2 a	7.9 a	52.4 a	53.3 a
T2-RT	67.3 a	8.0 a	52.9 a	53.0 a
T3-RB	66.9 a	7.5 a	52.5 a	52.8 a
T4-Ctrl	65.8 a	7.0 a	49.5 a	49.8 a
LSD	6.997	1.653	3.713	6.953

Means with a different letter(s) in each column are significantly different (LSD test, $p < 0.05$)

As previously observed, the effect of biostimulants on chiltepín development was most evident during the first 30 DAT, with treatments T1-BF, T2-RT, and T3-RB standing out in order of importance. The effect of T1-BF can be attributed to its components (Table 2). Exuroot stimulates root exudates, which strengthen the roots, allowing for prolonged water and nutrient absorption. Additionally, its biological composition enhances nutrient uptake efficiency, optimizing crop development.

T2-RT, which contains Exuroot along with a mix of *Bacillus subtilis* and *Trichoderma harzianum*, may serve a dual function. On one hand, it contributes to root nutrition, stimulation, and protection, and on the other, it helps prevent diseases caused by root pathogens such as *Phymatotrichopsis omnivora, Phytophthora spp., Rosellinia spp., Fusarium spp.*, and *Streptomyces scabies*. Its biological activity promotes both root development and overall crop growth.

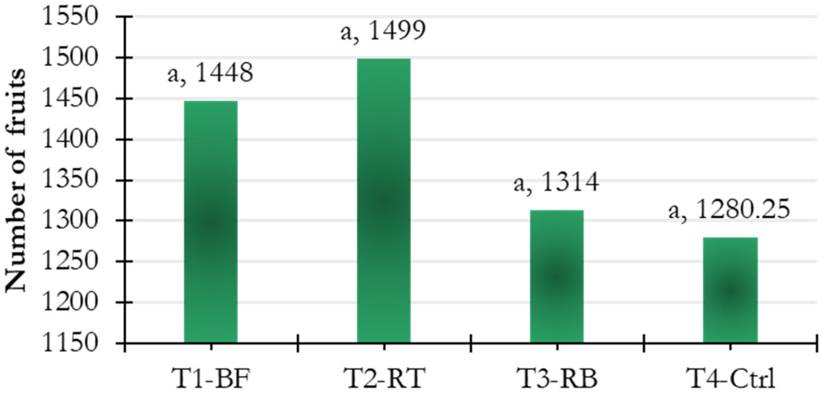
Finally, T3-RB, which contains beneficial fungi and bacteria, has demonstrated effectiveness against specific phytopathogens such as *Phytophthora capsici, Fusarium oxysporum, Pythium spp., Rhizoctonia spp.*, and *Verticillium spp. Bacillus* species are notable for their ability to compete for space and nutrients, as well as their production of antibiotic compounds that inhibit the growth of pathogenic fungi.

Other studies have shown that inoculation of chili plants with *Bacillus subtilis* and *Trichoderma harzianum* enhances plant height and root volume development (Gamboa-Angulo *et al.*, 2020). Additionally, research has indicated that inoculation with mycorrhizae and rhizobacteria improves chili plant growth (Aguirre-Medina and Espinosa, 2016). A bioformulation of *T. harzianum* using a substrate derived from a blend of wheat bran and peat has also shown promising results in promoting chili plant growth and photosynthetic capacity (Bader *et al.*, 2020). The use of beneficial microorganisms and root stimulators in agriculture presents a viable alternative for achieving improved crop performance.

Effect of biostimulants on chiltepín production and quality

The results for production variables, including the number of fruits, fresh weight, and dry weight of chiltepín, are presented in Figure 4. Significant differences ($p < 0.05$) were observed among treatments for fresh and dry weight, while no statistical differences were detected for the number of fruits. However, treatments T1-BF and T2-RT had the highest values for this variable. Similarly, fresh and dry weight were highest in treatments T1-BF and T2-RT, followed by T3-RB and T4-Ctrl, with the latter showing the lowest values.

The results for quality variables, such as fruit length (L), width (W), and the L/W ratio of chiltepín, are presented in Figure 5. Significant differences ($p < 0.05$) were observed among treatments for all three variables. Treatment T2-RT on fruit length showed the highest values, followed by T1-BF, T3-RB, and T4-Ctrl, with the control treatment showing the lowest value. Treatment T3-RB on fruit width achieved the highest values, followed by T2-RT and T1-BF, with the control treatment (T4-Ctrl) again registering the lowest value. Regarding the L/W ratio, treatment T2-RT reflected the highest values compared to the other treatments ($p = 0.05$).



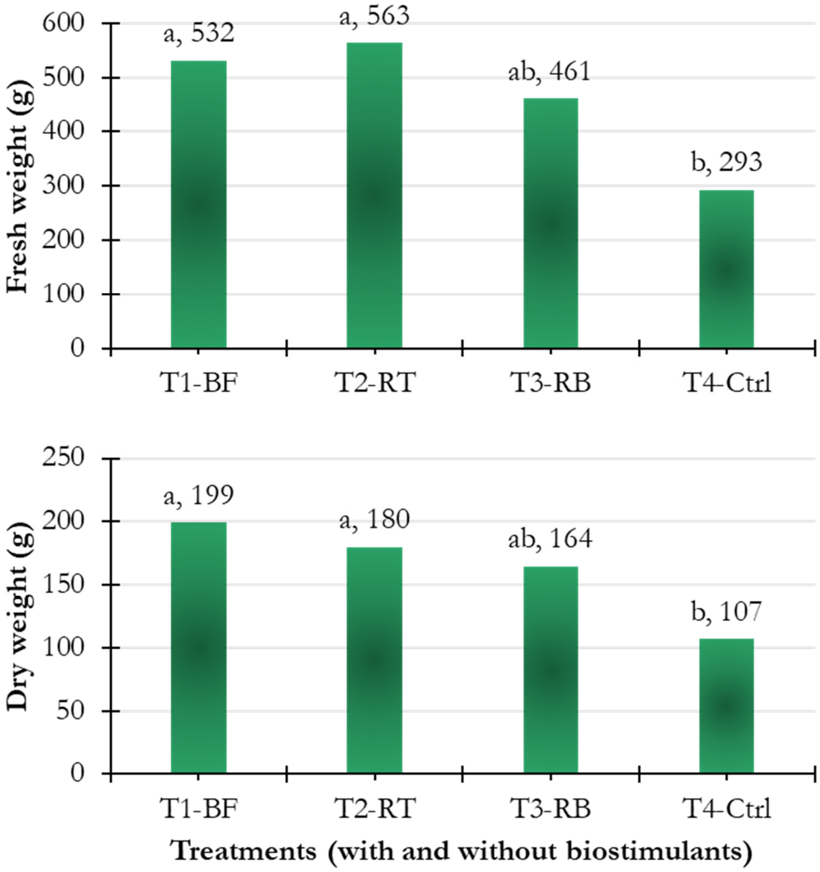
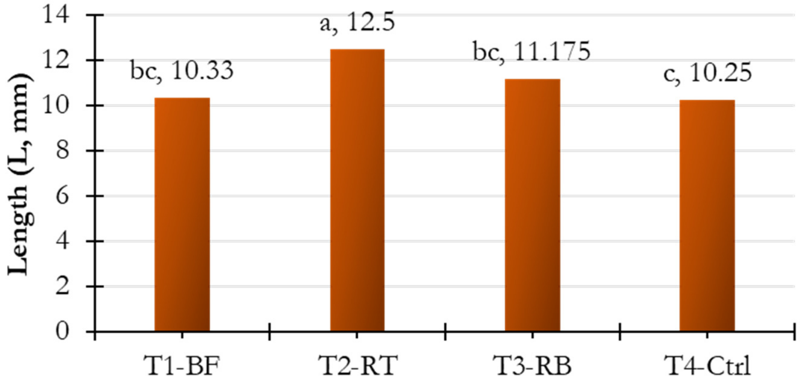


Figure 4. Effect of biostimulants on production variables: number of fruits (LSD = 643.41), fresh weight (LSD = 173.03), and dry weight (LSD = 62.2) of chiltepín using an agroecological system
Means with a different letter(s) above each bar are significantly different (LSD test, $p < 0.05$)



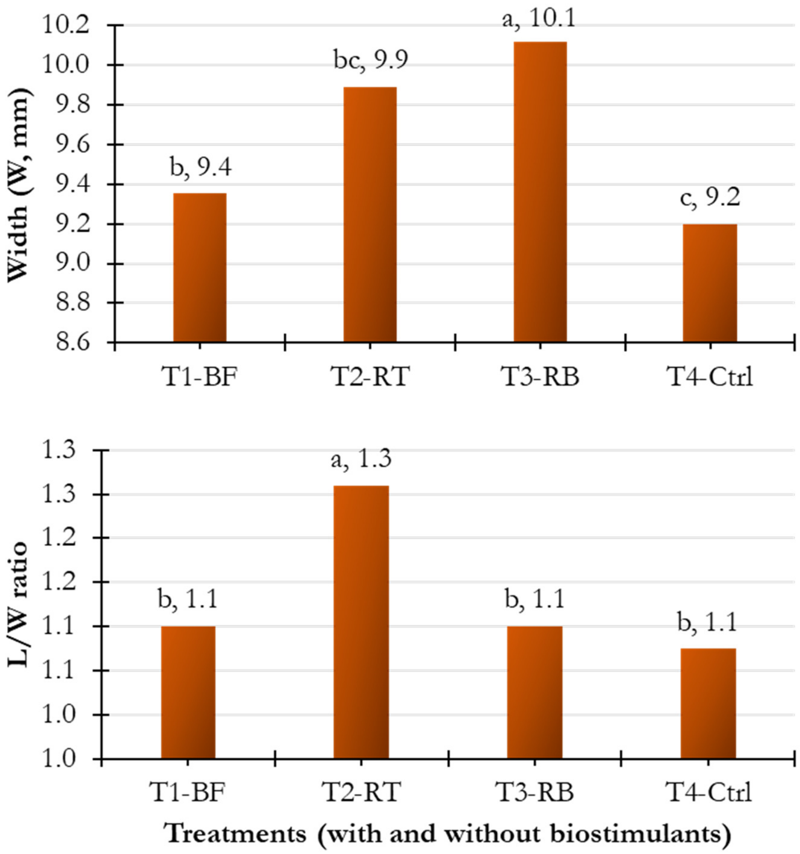


Figure 5. Effect of biostimulants on quality variables: fruit length (LSD = 0.979), width (LSD = 0.482), and L/W ratio (LSD = 0.1036) of chiltepín using an agroecological system
Means with a different letter(s) above each bar are significantly different (LSD test, $p < 0.05$)

Chiltepín is a wild chili species whose agronomic evaluation is based on the analysis of various fruit parameters, such as fresh and dry weight, length (L), width (W), and the L/W ratio (Figure 6). These parameters not only indicate quality and production, but are also crucial for selecting genotypes with superior agronomic traits and adaptability to diverse environmental conditions. In this context, the results of this study are significant and can be linked to the specific formulations used in the treatments.

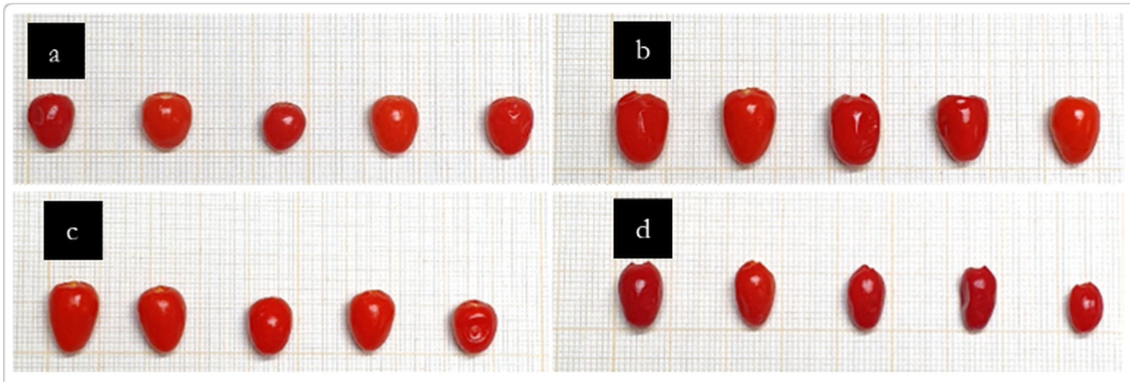


Figure 6. Effect of biostimulants on chiltepín fruit size using an agroecological system
(a) T1-BF; (b) T2-RT; (c) T3-RB; (d) T4-Ctrl

These findings provide valuable insights into the agronomic potential of chiltepín, offering responses to environmental stress and contributing to the crop's improvement. Various studies report strategies to achieve improvements, such as using biostimulation with beneficial microorganisms to optimize fruit production and quality, which are critical for commercialization. Aguirre-Medina and Espinosa (2016) mention that seed inoculation with mycorrhizal fungi and rhizobacteria improved plant growth and fruit production in jalapeño peppers. Another study indicates that beneficial microorganisms, when forming a consortium, improved the nutritional quality of chili fruits by increasing carotenoid content and antioxidant activity, which are important marketing traits (Sánchez *et al.*, 2022). Other researchers suggest that using vermicompost as a substrate enhances seed germination, plant growth, and crop production, with the best results observed in treatments containing 75% or more vermicompost (Palma-López *et al.*, 2020). Inoculation of chiltepín plants with *Glomus intraradices* and *Glomus fasciculatum* increased production by 135% compared to untreated plants (Mares-Quiñones and Valiente-Banuet, 2019). In this study, we observed a 47% increase in fresh fruit weight and a 46% increase in dry fruit weight compared to the control. Similarly, Ramírez *et al.* (2018) reported an average fruit width of 1.0 cm and a length of 0.93 cm. Our results show similarities in width with an average of 1.0 cm, but an improvement in fruit length with an average of 1.14 cm. Based on the agronomic variables, biostimulants had a significant effect on production and quality of chiltepín.

Conclusions

Morphological analyses revealed notable phenotypic variability between wild and domesticated varieties of chiltepín, highlighting the ecological adaptations and selective pressures that have shaped their evolution. This phenotypic diversity emphasizes the potential for chiltepín to respond to agricultural practices that promote its production.

The use of biostimulants, especially in treatments T1-BF and T2-RT, proved to be an effective tool for enhancing growth and development outside of the natural environmental and ecological conditions for this chili variety. These treatments significantly increased key agronomic parameters, such as height, stem diameter, chlorophyll content, and plant canopy, as well as nearly a 50% increase in the fresh and dry weight of the fruits. Additionally, an improvement in fruit quality was observed. Biostimulants were crucial in the plant's development and fruiting phases, highlighting their potential in chiltepín production using an agroecological approach.

Authors' Contributions

KCIL, LRH, and ACGF conceived idea and designed the research work. KCIL, LRH, and ACGF conducted the research, performed statistical analysis, and wrote the original draft. RIR, MCEDG, and JHH supervised the experimental work, critically reviewed and edited the document.

All authors read and approved the final manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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