

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

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ABSTRACT: Curly chili (*Capsicum annuum* L.) are a strategic horticultural commodity in Indonesia, but their productivity continues to fluctuate due to environmental stress and suboptimal cultivation techniques. To address these fluctuations, innovations in cultivation technology are needed, such as seed priming and the application of biostimulants based on bioactive peptides. This study aims to test and analyze the effects of seed priming and bioactive peptide concentration on vegetative growth variables, yield components, and fruit quality of the Hot Star F1 curly chili pepper variety. The study was conducted on farmland in Bangli Village, Baturiti Subdistrict, Tabanan, Bali, from September 2025 to April 2026. The experiment used a split-plot randomized block design (RBD) with three replications, in which the seed soaking method (B) was assigned as the subplot (consisting of treatment levels Bp, Bg, and Bc) and the peptide concentration (P) as the main plot (consisting of treatment levels 0, 2, and 4 mL L⁻¹). The data were analyzed using Analysis of Variance (ANOVA) and followed by the Least Significant Difference (LSD) test at the 5% level. The results of the study showed that peptide concentration had a highly significant effect on vegetative growth and fruit yield components, whereas the seed soaking method and its interactions had no significant effect. A peptide concentration of 2 mL L⁻¹ (P2) yielded the best results, with the highest stem diameter (8.51 mm), fruit length (12.04 cm), fruit diameter (7.05 mm), and number of fruits per plant, reaching 116.11 fruits. Conversely, the application of a high-concentration peptide solution at 4 mL L⁻¹ (P4) stimulated excessive vegetative growth (plant height of 47.01 cm and root dry weight of 9.94 g) but drastically reduced fruit productivity to only 47.22 fruits per plant due to an imbalance in assimilate allocation. This study concludes that the periodic application of bioactive peptides at a moderate concentration of 2 mL L⁻¹ (P2) is recommended to increase the productivity of curly chili peppers without compromising the plants' physiological balance.

KEYWORDS: Bioactive peptides, Curly chili peppers, Productivity, Seed soaking, Seed priming

I. INTRODUCTION

Curly chili (*Capsicum annuum* L.) are one of the leading horticultural commodities with high economic value and are a staple for the Indonesian people. Demand for curly chili peppers tends to increase every year, both for household consumption and the food industry, making them crucial to national food security (Central Bureau of Statistics, 2023). However, the availability of curly chili peppers in the market often fluctuates due to variations in production at the farmer level. According to data from the Central Statistics Agency (2023), over the past four years (2021–2024), national curly chili production has shown an unstable pattern, rising from 8.60 million kg in 2021 to 11.59 million kg in 2023, but then dropping sharply to 10.85 million kg in 2024 due to climate anomalies, pest infestations, and a shrinking harvest area (2023).

To address these production fluctuations, innovations in cultivation technology are needed, starting with the preparation of high-quality planting materials. Seed priming is a pretreatment technology that effectively improves germination quality, accelerates early vigor, and stimulates the plant's initial physiological response through the processes of seed imbibition and enzymatic activation (Meo et al., 2024; Pompelli et al., 2023). Metode priming, seperti hidropriming, osmopriming, dan biopriming dengan berbagai bahan, terbukti meningkatkan ketahanan bibit terhadap cekaman lingkungan (Granata et al., 2024; Koushal et al., 2025). Selain optimasi benih, peningkatan produktivitas cabai keriting juga memerlukan inovasi dalam aplikasi senyawa bioaktif yang dapat merangsang pertumbuhan tanaman (Nuni et al., 2022).

In addition to seed treatment, the introduction of exogenous bioactive compounds, such as bioactive peptides, is a promising modern technology for stimulating sustainable plant growth (Prasad, 2024). Bioactive peptides act as signaling molecules capable

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

of activating growth transduction pathways at the cellular level, increasing the expression of stress-tolerance-related genes, and optimizing photosynthetic activity and plant cell division (Ren et al., 2024; Zhang, 2025).

Although the effects of seed priming technology and the application of bioactive peptides have been studied separately by several previous researchers, studies on the interaction between these two technological approaches in horticultural crops particularly curly chili peppers remain very limited. Understanding how the physiological response to germination induced by priming interacts with bioactive peptide stimulation during the vegetative and generative phases in the field remains an important research gap that needs to be addressed. Therefore, this study aims to test, analyze, and determine the effect of the interaction between the seed priming method and the concentration level of bioactive peptides on vegetative growth variables, yield components, and fruit quality of curly chili plants. Through this study, it is hoped that practical technical recommendations can be formulated to address production constraints in curly chili cultivation and efficiently achieve the best possible harvest.

II. RESEARCH METHODOLOGY

Research Design

The study employed a randomized block design (RBD) with a split-plot arrangement, involving 2 factors and 3 replications. The first factor, as a subplot, was the seed soaking method prior to sowing (B), which consisted of 3 levels, namely:

1. Bp = Seeds were soaked for 12 hours following the farmers' method, using warm water with an initial temperature of 50°C, with the temperature not maintained at a constant level.
2. Bg = Seeds were soaked for 12 hours in warm water with an initial temperature of 50°C (temperature not maintained at a constant level) supplemented with 1 mL L⁻¹ of Golden Grow (GG) and 2 mL L⁻¹ of peptide.
3. Bc = Seeds were soaked in tap water with 1 mL L⁻¹ of Golden Grow (GG) and 2 mL L⁻¹ of peptide for 12 hours.

The second main factor is peptide concentration (P), which consists of three levels, namely:

1. P0 = peptide concentration of 0 mL L⁻¹ of water (no peptide)
2. P2 = peptide concentration of 2 mL L⁻¹ of water
3. P4 = peptide concentration of 4 mL L⁻¹ of water

Thus, there are 9 treatment combinations (P0Bp, P0Bg, P0Bc, P2Bp, P2Bg, P2Bc, P4Bp, P4Bg, P4Bc) with 3 replicates, requiring a total of 27 experimental plots.

Table 1. Illustration of the arrangement of curly chili seeds after soaking/marinating using methods Bp, Bg, and Bc

peptide concentration of 0 mL L ⁻¹ of water (P0)			peptide concentration of 2 mL L ⁻¹ of water (P2)			peptide concentration of 4 mL L ⁻¹ of water (P4)		
I	II	III	I	II	III	I	II	III
P0Bp	P0Bg	P0Bc	P2Bg	P2Bp	P2Bc	P4Bg	P4Bp	P4Bc
P0Bg	P0Bc	P0Bp	P2Bp	P2Bc	P2Bg	P4Bc	P4Bg	P4Bp
P0Bc	P0Bp	P0Bg	P2Bc	P2Bg	P2Bp	P4Bp	P4Bg	P4Bc

Research Location and Time

The research was conducted from September 2025 to March 2026 on farmland in Bangli Village, Baturiti Subdistrict, Tabanan Regency, Bali Province. Geographically, the research site is located at coordinates 8°19'30" S and 115°10'30" E (WGS84 Coordinate System) at an elevation of approximately 1,200 m above sea level. The soil type at the research site is Andisol. Climate conditions at the site during the study period included a temperature of 28°C and humidity of 65.75% for the highland region of Bali during that season. The site's previous land-use history consisted of intensive horticultural farming (Figure 1).

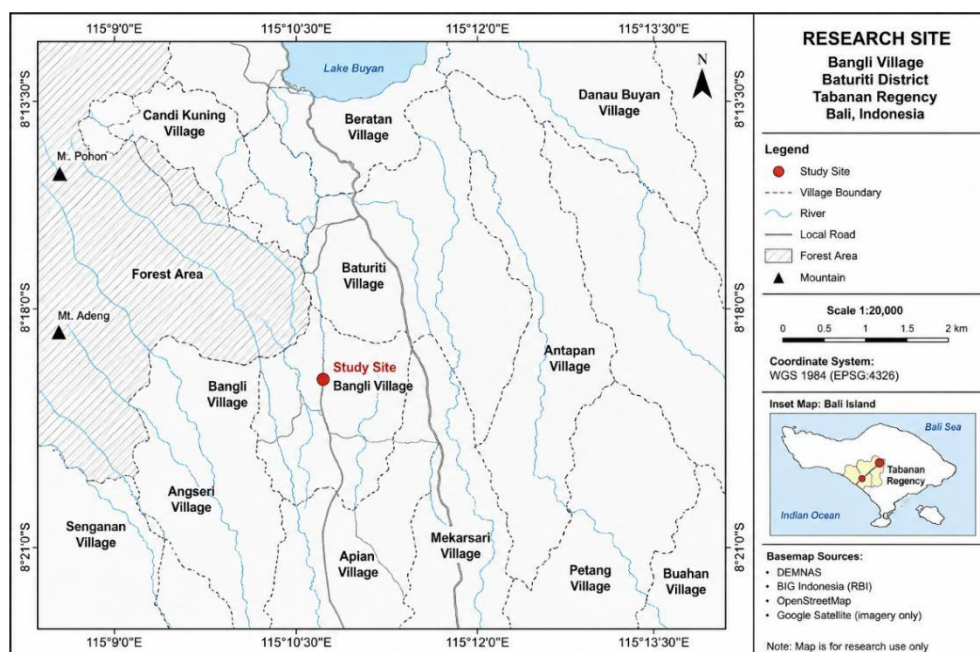


Figure 1. Geographic location of the experimental field in Bangli Village, Baturiti District, Tabanan Regency, Bali, Indonesia.
The map was prepared using WGS 1984 (EPSG:4326) with DEMNAS and OpenStreetMap as basemap sources.

Research Materials and Equipment

The materials used included Hot Star F1 curly chili pepper (*Capsicum annuum* L.) seeds, water, Golden Grow, a peptide-based biostimulant, organic fertilizer, NPK fertilizer, dolomite, botanical pesticides, and plastic mulch. Samples of chili plants, roots, foliage, and fruits were used for observation and measurement during the study. The roots were washed with clean water before being dried in an oven until they reached a constant weight. The equipment used in the study included a measuring tape, a ruler, a digital caliper, digital and analytical scales, a drying oven, pruning shears, a knife, trays, buckets, a sprayer, paper bags, labels, writing utensils, and a camera for documentation.

Research Variables

The variables observed include growth parameters, yield, and yield quality:

- 1) Plant Height (cm): Measured from the base of the stem to the tip of the apical meristem using a measuring tape or long ruler on labeled curly chili pepper plant samples; recorded periodically by hand on observation sheets every 2 weeks starting 14 days after planting (DAP) until harvest.
- 2) Stem Diameter (mm): Measured on the stem of the curly chili plant (2 cm above the soil surface) using a digital caliper once every 2 weeks starting at 14 days after sowing.
- 3) Fresh and Dry Weights of the Plant's Above-Ground Parts and Roots (g): Measurements were taken at harvest by pulling up the entire plant, then separating the roots from the above-ground parts using pruning shears or a knife. The root sections were carefully washed on a tray under running water until completely free of soil. The root and shoot samples were then placed in labeled paper bags, dried in a drying oven until they reached a constant weight, and weighed on an analytical balance.
- 4) Number of Fruits per Plant (fruits): Counted manually from all chili peppers formed on the sample plants using a manual counter and recorded directly in the observation logbook at each harvest period.
- 5) Fruit Weight per Plant (g): Weighed using a digital scale at each harvest (3 harvests).
- 6) Fruit Length (cm) and Diameter (mm): Measured on representative chili pepper samples using a digital caliper or ruler for fruit length, and a digital caliper for fruit diameter.
- 7) Fruit Flesh Thickness (mm): Measured on chili pepper samples that have been cut crosswise using a digital caliper to determine the thickness of the fruit flesh.
- 8) Fruit diameter (mm): Measured at harvest using digital calipers at the widest point of the fruit.

Research Procedure

- 1) Seed Soaking Curly chili seeds were divided into three lots: Bp (soaked in warm water at 50°C temperature not maintained at a constant level for 12 hours, following the method used by local farmers), Bg (same treatment as Bp, but the soaking water was supplemented with 1 mL L⁻¹ of Golden Grow/GG and 2 mL L⁻¹ of peptide), and Bc (soaked in tap

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

water [room temperature] for 12 hours with the addition of 1 mL L⁻¹ of GG and 2 mL L⁻¹ of peptide). Soaking was carried out simultaneously; afterward, the seeds were drained and air-dried in a cool place.

- 2) Sowing and Seedling Rearing: Seeds are sown one per hole in a tray filled with a growing medium mixture of fine soil, rice husk charcoal, cow manure, and dolomite (2:1:1:1). Seedlings are sprayed with GG Daun at a concentration of 1 mL L⁻¹ at 7 and 14 days after being transplanted into polybags (applying the solution to the foliage and growing medium). Seedlings are ready for planting at 20–25 days old with at least 5 fully developed leaves; three days before transplanting, the foliage is sprayed evenly with 2 mL L⁻¹ of peptide.
- 3) Soil Preparation and Bed Construction Concurrent with seed sowing, three plots of relatively similar size were selected for the main treatments (P0, P1, P2), with homogeneous conditions (soil type, fertility, sunlight, irrigation source, slope), and ideally from a single landowner to ensure a uniform land-use history. The treatment positions were randomized and coded. The field is cleared of plant debris and weeds, tilled (to a depth of 25–30 cm) using a mini-tractor in accordance with local farming practices, fertilized with a base fertilizer (mature organic fertilizer + NPK) 7–10 days before planting, and then leveled. Raised beds are made 100–120 cm wide, covered with silver-black mulch, and planted using a double-row system with planting holes spaced 50×60 cm apart.
- 4) Mulch Installation, Planting, and Fertilization: Make holes in the mulch according to the planting spacing. Planting should be done in the morning or evening to prevent wilting. Select uniform seedlings, remove them from the trays, plant them up to the root collar, firm the soil, and then water sufficiently. Fertilizer should be applied at the recommended dosage using the hole-planting method, with holes spaced 10 cm from the base of the stem.
- 5) Staking and Maintenance Bamboo stakes (4×100 cm) are installed immediately after planting, 10 cm from the plant and buried 15–20 cm deep. Tie the plants with twine at 30–40 days after planting (DAP) or when the first branch appears. Prune (wiwil) the axillary buds at 10–12 DAP. Pest control prioritizes environmentally friendly practices using the botanical pesticide Bumbu Bali (250 mL L⁻¹ of water); synthetic pesticides may be combined according to the recommended dosage in cases of severe pest infestation.
- 6) Application of Peptide and Golden Grow (GG) in the Field: Peptide was sprayed evenly onto the shoots every week, starting at 7 days after planting (DAP) until the end of the study. GG was applied via soil drenching at a rate of 1 mL per liter of water every week starting 1 week after planting, and application was discontinued when the plants began to flower. GG application was resumed two weeks after peak harvest to stimulate a return to optimal production.
- 7) Harvesting and Post-Harvest Handling Harvesting is conducted at ±85–100 days after sowing (DAS): early harvest (±85–90 DAS) when the fruit is ±80% ripe (predominantly red with a hint of green), optimal harvest (±90–100 DAS) when the fruit is bright red, and subsequent harvests every 3–7 days depending on plant condition. Fruits are handpicked along with their stems to prevent damage, then sorted to remove fruits infested with pests or damaged fruits, and finally stored in a dry, well-ventilated area to maintain quality and shelf life.

Data Analysis

Data analysis was performed using Analysis of Variance (ANOVA). If a treatment showed a significant interaction effect ($P < 0.05$), it was followed by Duncan's Multiple Range Test (DMRT) at the 5% level. If only a single factor has a significant effect, a Least Significant Difference (LSD) test at the 5% level is conducted to determine the differences between treatments.

III. RESULTS

Significance of the Effects of Treatments on Observation Variables

Table 2 shows the significance of the seed soaking treatment (B), the peptide concentration treatment (P), and the interaction effect of the two treatments ($B \times P$) on various observation variables. Based on the analysis of variance, the interaction between seed soaking and peptide concentration ($B \times P$) had no significant effect on any of the observed variables ($P \geq 0.05$).

The results of the analysis of variance showed that seed soaking (B) had no significant effect on any of the observed variables, namely plant height, stem diameter, dry weight of the canopy, dry weight of the roots, fruit length, number of fruits per plant, fruit diameter, and fruit flesh thickness ($P \geq 0.05$). Peptide concentration (P) had a highly significant effect on plant height, root dry weight, number of fruits per plant, and fruit diameter ($P < 0.01$), as well as a significant effect on stem diameter ($P < 0.05$). However, peptide concentration had no significant effect on canopy dry weight, fruit length, and fruit flesh thickness ($P \geq 0.05$).

The results of the analysis of variance showed that the single treatment of seed soaking (B) had no significant effect on any of the observed variables, including plant height, stem diameter, dry weight of the canopy, dry weight of the roots, fruit length, number of fruits per plant, fruit diameter, and fruit flesh thickness ($P \geq 0.05$). In contrast, the single treatment of peptide concentration (P) had different effects on several observed variables. Peptide concentration had a highly significant effect on plant

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

height, root dry weight, number of fruits per plant, and fruit diameter ($P < 0.01$), as well as a significant effect on stem diameter ($P < 0.05$). However, peptide concentration had no significant effect on canopy dry weight, fruit length, and fruit flesh thickness ($P \geq 0.05$).

Table 2. Significance of the Effects of Seed Soaking (B), Peptide Concentration (P), and Interaction (B × P) on the Observed Variables.

No.	Variable	Treatment		
		B	P	B × P
1.	Plant Height (cm)	ns	**	ns
2.	Stem Diameter (mm)	ns	*	ns
3.	Dry Weight of the Crown	ns	Ns	ns
4.	Dry Weight of Roots	ns	**	ns
5.	Fruit Length (cm)	ns	Ns	ns
6.	Number of Fruits per Plant	ns	**	ns
7.	Fruit Diameter (mm)	ns	**	ns
8.	Fruit Flesh Thickness (mm)	ns	ns	ns

Notes:

Ns : No Significant Effect ($P \geq 0.05$)

* : Significant Effect ($P < 0.05$)

** : Highly Significant Effect ($P < 0.01$)

Td : Not Statistically Analyzed

Vegetative Growth of Curly Chili Peppers

Based on the analysis of variance, the single treatment of peptide concentration (P) had a highly significant effect on plant height, stem diameter, and dry root weight. In contrast, the seed soaking method (B) and its interaction with peptide concentration (B×P) had no significant effect on the vegetative growth of the plants (Table 3).

Table 3. Plant Height, Stem Diameter, Dry Weight Of The Canopy, and Dry Weight of The Roots of Curly Chili Pepper Plants Under Various Treatments

Treatment	Plant Height (cm)	Stem Diameter (mm)	Dry Weight of Canopy (g)	Dry Weight of Roots (g)
Seed Soaking (B)				
Bp	43.32 b	7.82 a	23.33 a	8.44 a
Bg	44.94 a	8.09 a	23.44 a	7.83 a
Bc	43.29 b	7.83 a	23.11 a	8.06 a
BNT 5%	1.60	0.48	3.14	3.34
Peptide Concentration (P)				
P0 (0 mL L ⁻¹)	40.88 c	7.42 b	21.67 b	5.72 c
P2 (2 mL L ⁻¹)	43.66 b	8.51 a	23.11 ab	8.67 b
P4 (4 mL L ⁻¹)	47.01 a	7.82 b	25.11 a	9.94 a
BNT 5%	2.01	0.50	2.27	0.69

Note: Numbers followed by the same letter in the same column indicate no significant difference based on the BNT test at the 5% level.

The results of the observations showed that higher peptide concentrations significantly stimulated plant height and root dry weight. The highest average plant height was achieved by treatment P4 (47.01 cm), which differed significantly from P2 (43.66 cm) and P0 (40.88 cm). Similarly, for the dry root weight parameter, increasing peptide concentrations resulted in significantly different weights at each level, with the highest value observed at P4 (9.94 g), followed by P2 (8.67 g), and the lowest at the control P0 (5.72 g). However, the stem diameter response showed an interesting quadratic pattern, with the highest value actually achieved at the moderate concentration of P2 (8.51 mm) and a significant decrease observed at the high concentration of P4 (7.82 mm).

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

Yield and Quality of Curly Chili Peppers

Similar to the vegetative growth parameters, an analysis of variance of the yield and fruit quality components revealed a highly significant effect of the single treatment of peptide concentration (P). The seed soaking method (B) and its interaction (BxP) had no significant effect on any of the observed fruit parameters (Table 4).

Table 4. Fruit Length, Number of Fruits, Fruit Diameter, and Fruit Flesh Thickness of Curly Chili Pepper Plants Under Various Treatments

Treatment	Fruit Length (cm)	Number of Fruits per Plant	Fruit Diameter (mm)	Flesh Thickness (mm)
Seed Soaking (B)				
Bp	11.14 a	73.78 a	6.30 a	0.76 a
Bg	11.26 a	71.62 a	6.54 a	0.84 a
Bc	10.55 a	78.16 a	6.44 a	0.77 a
BNT 5%	0.98	18.06	0.38	0.09
Peptide Concentration (P)				
P0 (0 mL L ⁻¹)	10.11 b	60.22 b	5.71 c	0.76 a
P2 (2 mL L ⁻¹)	12.04 a	116.11 a	7.05 a	0.84 a
P4 (4 mL L ⁻¹)	10.80 ab	47.22 c	6.51 b	0.77 a
BNT 5%	1.24	10.42	0.42	0.14

Note: Numbers followed by the same letter in the same column indicate no significant difference based on the BNT test at the 5% level.

Further analysis of the 5% BNT treatment on fruit components showed the exceptional dominance of the medium-concentration peptide treatment P2 (2 mL L⁻¹). The P2 treatment significantly produced the best yield, with the number of fruits per plant reaching 116.11, far exceeding the control P0 (60.22 fruits) and the high-dose treatment P4 (47.22 fruits). Fruit quality parameters also showed a similar pattern, with maximum fruit length observed in P2 (12.04 cm) which was significantly different from P0 (10.11 cm) and the largest fruit diameter also observed in P2 (7.05 mm), which was significantly different from both P4 (6.51 mm) and P0 (5.71 mm). Interestingly, fruit flesh thickness was not significantly affected by any of the treatments, with averages ranging from 0.76 mm to 0.84 mm.

IV. DISCUSSION

The application of bioactive peptides has been shown to trigger a strong growth response in curly chili peppers. Biochemically, bioactive peptides act as signaling molecules that activate plant hormone signal transduction pathways, modulate gene expression for stress tolerance, and accelerate cell division (Zhang, 2025; Prasad, 2024). The linear increase in plant height up to a concentration of 4 mL L⁻¹ (P4) indicates highly intensive stimulation of cell division in the apical meristem due to the supply of amino acids and these external molecular signals.

The very strong vegetative stimulation resulting from peptide application is also reflected in the root dry weight parameter. Plant roots underwent massive growth and structural expansion as the peptide dose increased (reaching 9.94 g at P4). Physiologically, robust root growth maximizes the capacity to absorb nutrients and water to supply the plant's aboveground parts, expands the root system's reach, and facilitates more efficient nutrient assimilation to support the plant's overall metabolism (Abinandan et al., 2024).

Although vegetative growth (plant height and root dry weight) increased linearly up to the highest dose, P4, the plant productivity response (number of fruits per plant) instead showed a contrasting nonlinear (quadratic) pattern. Treatment P2 (2 mL L⁻¹) yielded the highest fruit count of 116.11 fruits per plant, while the high dose of P4 (4 mL L⁻¹) actually experienced a drastic decrease to 47.22 fruits, which was even lower than the control without peptides (P0; 60.22 fruits). This phenomenon can be explained by the theory of assimilate allocation (sink-source relationship) and plant hormonal regulation. When an excessive concentration of the peptide was applied (P4), overstimulation occurred in vegetative organs such as the leaves (canopy) and roots, causing the plants to prioritize the allocation of photosynthates toward stem cell elongation and root biomass growth (as indicated by the highest canopy dry weight of 25.11 g and the highest root dry weight of 9.94 g at P4). As a result, the energy and assimilates that should be allocated for the initiation of flowering and fruit development become severely limited, leading to flower drop or suboptimal fruit set. In contrast, at a moderate concentration of P2 (2 mL L⁻¹), an ideal balance (physiological balance) is achieved between vegetative and reproductive growth. The sturdy stem structure (maximum stem diameter of 8.51

The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

mm) of P2 enables optimal transport of nutrients and water to support the formation of a large number of fruits (116.11 fruits), with the best length (12.04 cm) and diameter (7.05 mm).

On the other hand, the lack of a significant effect of the seed soaking method (B) on nearly all growth and yield variables indicates that seed priming serves only as a physiological stimulant during the early stages of germination (imbibition, activation of amylase/protease enzymes, and dormancy breaking) (Koushal et al., 2025; Pompelli et al., 2023). Once the plants have been transplanted to the field and receive intensive foliar fertilizer applications as well as continuous weekly peptide sprays, the initial physiological effects of this seed priming are reduced and masked by the dominance of nutrients and plant growth regulators during the field's vegetative-generative phase.

V. CONCLUSION

This study concludes that the application of bioactive peptides significantly affects the vegetative growth and yield components of the Hot Star F1 curly chili pepper variety, whereas neither the seed priming method nor the interaction between these two factors had a significant effect. A peptide concentration of 2 mL L⁻¹ (P2) proved to be the best treatment because it created an ideal physiological balance for the plants, resulting in the largest stem diameter (8.51 mm), maximum fruit size (12.04 cm in length and 7.05 mm in diameter), and the highest productivity, reaching 116.11 fruits per plant. Conversely, the application of a high-concentration peptide solution at 4 mL L⁻¹ (P4) actually triggered excessive vegetative growth (plant height of 47.01 cm and root dry weight of 9.94 g), which significantly and drastically reduced fruit yield to only 47.22 fruits per plant due to an imbalance in the allocation of assimilate energy. Therefore, to optimize the cultivation and yield of curly chili peppers in a sustainable manner, farmers are strongly encouraged to apply bioactive peptides periodically at a moderate concentration of 2 mL L⁻¹ (P2).

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The Effect of Seed Soaking and Peptide Concentration on the Growth and Yield of Curly Chili (*Capsicum annuum* L.)

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