

# The Effect of Seed Sowing Density on Growth Parameters in Six Different Microgreen

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#### ABSTRACT

Seed sowing density is a key parameter that directly affects plant growth and the final product quality in microgreens. Proper adjustment of sowing density ensures optimal growth conditions, enhancing yield and supporting healthy and vigorous plant development. In this study, broccoli, black radish, red beet, pea, sunflower, and bean seeds were used: three different sowing densities were determined, and the seeds were sown in containers measuring 16x9x7 cm. The study investigates the effects of different seed sowing densities on plant height, hypocotyl length, stem diameter, individual plant weight, leaf area, yield, dry matter content, and chlorophyll content in microgreens. Significant changes in plant growth parameters were observed as the sowing density increased. These findings highlight the necessity of carefully optimizing sowing density in microgreens production.

#### Plant Physiology

#### **Research Article**

Article History	
Received	: 28.10.2024
Accepted	: 22.03.2025

#### Keywords

Microgreens Seed sowing density Physical parameter

Altı Farklı Mikroyeşillikte Ekim Yoğunluğunun Büyüme Parametreleri Üzerine Etkisi

#### ÖZET

Mikroyeşilliklerde tohum ekim sıklığı, bitki gelişimini ve nihai ürün kalitesini doğrudan etkileyen temel bir parametredir. Tohumların ekim sıklığının doğru ayarlanması, optimal büyüme koşullarını sağlayarak verimi artırıp bitkilerin sağlıklı ve güçlü gelişimini destekler. Bu çalışmada, brokoli, siyah turp, kırmızı pancar, bezelye, ayçiçeği ve fasulye tohumları kullanılmış; üç farklı ekim sıklığı belirlenmiş ve 16x9x7 cm boyutlarındaki kaplarda tohum ekimi gerçekleştirilmiştir. Calışma, farklı tohum ekim sıklığının mikroyeşilliklerde bitki boyu, hipokotil boyu, gövde çapı, tek bitki ağırlığı, yaprak alanı, verim, kuru madde ve krolofil içeriği üzerine etkilerini ortava kovmaktadır. Tohum ekim sıklığı arttıkça bitki büyüme parametrelerinde önemli değisiklikler olduğu gözlemlenmiştir. Bu bulgular, mikroyeşillik üretiminde tohum ekim sıklığının dikkatli bir şekilde optimize edilmesi gerektiğini vurgulamaktadır.

Bitki Fizyolojisi

Araştırma Makalesi

Makale TarihçesiGeliş Tarihi28.10.2024Kabul Tarihi22.03.2025

Anahtar Kelimeler Mikro yeşillik Tohum yoğunluğu Fiziksel parametreler

Atıf İçin : Balık, S., & Yıldız-Daşgan, H. (2025). Altı Farklı Mikroyeşillikte Ekim Yoğunluğunun Büyüme Parametreleri Üzerine Etkisi.. *KSÜ Tarım ve Doğa Derg 28* (3), 661-671. DOI: 10.18016/ksutarimdoga.vi.1574906.
 To Cite: Balık, S., & Yıldız-Daşgan, H. (2025). The Effect of Seed Sowing Density on Growth Parameters in Six Different Microgreen.. *KSU J. Agric Nat 28* (3), 661-671. DOI: 10.18016/ksutarimdoga.vi.1574906.

## INTRODUCTION

It is estimated that approximately 2 billion individuals worldwide suffer from hidden hunger, a chronic deficiency in essential micronutrients such as vitamins and minerals (Lowe, 2021). In today's world, healthy nutrition is prioritized for the prevention of certain diseases. As global public health awareness increases, the demand for functional foods offering various health benefits is also rising (Yaşa et al., 2023). Vitamins are crucial micronutrients for human health, and deficiencies can lead to various illnesses. Therefore, in addition to being consumed through foods, they are also made available as supplements or incorporated into functional foods. (Saman & Tomaş, 2022). It has been reported that microgreens contain significantly higher levels of functional components, such as phenolic compounds and antioxidants, compared to the amounts found in mature leaves (Gök et al., 2024).

Microgreens have garnered substantial attention for their health and beauty benefits, boasting nutritional content

up to 40 times higher than mature vegetables. Their versatility in culinary applications and positive impact on environmental sustainability and economic viability further contribute to their popularity. Grown either in soil or hydroponically, microgreens are a leading crop in controlled environment agriculture. These young greens, harvested between the sprout and baby green stages, have gained widespread appeal due to rising public interest in healthy eating. Derived from herb, vegetable, and grain seeds, microgreens typically develop a central stalk with two sets of immature true leaves, following the emergence of cotyledons (Kyriacou et al., 2020; Chandrashekharaiah, 2013).

Microgreens production involves the use of dense sowing techniques, which increase competition for resources and consequently require higher seed quantities. Variations in seed size among vegetable species necessitate the adjustment of sowing density according to the specific species. Improper sowing density, whether excessive or insufficient, can lead to negative outcomes. In this context, preliminary research to determine the optimal seed quantities for different species is crucial (Sarıyer et al., 2024). Sowing density is a significant factor influencing microgreens yield; as sowing density increases, the weight of each plant decreases due to competition among plants, while the overall yield rises with the increased number of seeds per unit area. This increase continues until the maximum production capacity is reached (Thuong et al., 2020; Palmitessa et al., 2020; Moraru et al., 2022).

This study addresses significant issues in microgreens cultivation by focusing on the effects of seed sowing density on product quality, yield, and economic profitability. The amount of seeds used during cultivation significantly influences the physical characteristics, germination rate, and growth performance of microgreens. While higher seed sowing densities may enhance yield in the short term, they can also increase production costs, thereby reducing profit margins. Therefore, determining the optimal seed sowing density for each species is crucial for improving both growth quality and economic efficiency. This research aims to identify the ideal seed sowing density for selected microgreens species to optimize growth performance, minimize production costs, and maximize profitability. By investigating the effects of sowing density on physical characteristics and yield, this study seeks to contribute to the improvement of production processes and the provision of higher-quality products to consumers.

## MATERIAL and METHOD

## **Plant Material**

This study was conducted in the climate chamber of the Department of Horticultural Sciences, Faculty of Agriculture, Çukurova University. The seeds used in the study included black radish (*Raphanus sativus niger*) and broccoli (*Brassica oleracea italica*) from the *Brassicaceae* family, red beet (*Beta vulgaris cicla*) from the *Amaranthaceae* family, pea (*Pisum sativum*) from the *Leguminosae* family, sunflower (*Helianthus annuus*) from the *Asteraceae* family, and bean (*Phaseolus vulgaris*) from *the Leguminosae* family. The seeds were not subjected to any chemical treatment or pesticide application. The black radish, broccoli, and red beet seeds were obtained from the standard varieties of Arzuman Seed Company, the pea seeds from the standard variety of Intfa Company, the sunflower seeds from the standard variety of AGR Company, and the bean seeds from the germplasm bank of the Department of Horticultural Sciences.

## Plant Growing Conditions

The experiments were conducted in a climate-controlled growth chamber to determine the optimal seed sowing density for microgreens plants. Black radish, broccoli, and red beet were cultivated at 20 °C[Balik et al., 2024a], while pea, bean, and sunflower were grown at 23 °C, all under conditions of 50% relative humidity. The growth environment was supported by LED lamps designed to provide a balanced light spectrum similar to natural sunlight. These lamps ensured consistent light quality and intensity (350 µmol m-2 s-1), optimizing photosynthesis. Microgreens were subjected to 16-hour light and 8-hour dark cycles during cultivation. Transparent plastic containers measuring 16 cm × 9 cm × 7 cm (length × width × height) were used as growing trays, each filled with 250 cm<sup>3</sup> of peat. Experimental treatments were arranged in a completely randomized design, ensuring equal distribution across all parcels. This systematic approach aimed to enhance the accuracy and reliability of the research findings.

In this study, the selected plant species were sown at three different planting densities or seed rates based on seed size and weight (Table 1). The treatments for each plant species were as follows:

- 1. Treatment (Control): Seeds were sown to fully cover the surface area of the containers.
- 2. Treatment: Seeds were sown at 50% higher than the surface area capacity.
- 3. Treatment: Seeds were sown at 100% higher than the surface area capacity.

Çizelge 1. Her uygulama için belirlenen tohum ekim yoğunluğu seviyeleri (g)					
Plant species	Treatment 1	Treatment 2	Treatment 3		
Broccoli	13	19.5	26		
Black radish	32	48	64		
Red beet	16	24	32		
Pea	100	150	200		
Sunflower	28	42	56		
Bean	70	105	140		

Table 1. The seed sowing density levels established for each application (g)

## **Plant Nutrition**

The sown seeds were irrigated with pure water until the first green cotyledon leaves appeared. Immediately after the emergence of the green cotyledons, irrigation was performed with the nutrient solution specified below. Modified Hoagland nutrient solutions at half strength were used for microgreens production. The plants were supplied with the following nutrient solution at  $\frac{1}{4}$  strength (in mg/L): N (200), P (50), K (300), Ca (200), Mg (65), Fe (5.0), Mn (0.8), Cu (0.3), Zn (0.3), B (0.3), and Mo (0.05). The pH was set at 5.5, and the electrical conductivity (EC) was maintained between 1.2-1.6 dS cm<sup>-1</sup> (for both initial and later stages) during irrigation [Balik et al., 2024b].

## Plant Harvest

Microgreens were cultivated for a period of 7-15 days, depending on the growth rate of six different plant species. Harvesting was conducted when the seedlings had fully developed their first true leaf, the second leaf had begun to emerge, and the cotyledon leaves had reached the rounded margin stage.

## Measurements of Plant Growth Parameters

During the harvest, measurements of plant height, hypocotyl length, stem diameter, and individual plant weight were taken for microgreens from six different plant species, with four replicates and ten plants measured per replicate under various cultivation conditions. Yield per unit area was calculated. The leaf area per plant was determined using a leaf area meter (Li-3100, LICOR, Lincoln, NE, USA), expressed in square centimeters. After harvest, the plants were weighed using a digital scale to determine the fresh leaf weight per plant in grams. Chlorophyll content in the leaves was measured using a SPAD chlorophyll meter (Minolta 502, Osaka, Japan). The fresh leaves were subsequently dried in an oven at 65 °C for 48 hours, and the dry weight per plant was recorded.

## Statistical Analyses

All data were analyzed using JMP v5.0.1 statistical software, and an analysis of variance (ANOVA) was conducted. Treatment means were compared using LSD's significant difference test at  $p \le 0.05$ . The significance levels for the three-way ANOVA analyzing the effects of plant species, cultivation environment, and their interactions are presented in Table 2.

## RESULTS

Microgreens, a nutrient-rich source, exhibit varying growth patterns depending on the seed sowing density applied during cultivation. This study investigates the effects of three different sowing densities—Full Surface Coverage Sowing (T1), 50% above the surface area capacity (T2), and 100% above the surface area capacity (T3)—on plant height, hypocotyl length, stem diameter, single plant weight, leaf area, yield, dry matter ratio, and SPAD-chlorophyll content across six microgreens species: broccoli, black radish, red beet, pea, sunflower, and bean (Table 3).

## Broccoli

In broccoli microgreens, under Full Surface Coverage Sowing (T1) conditions, plant height was measured at 6.81 cm, hypocotyl length at 5.03 cm, and stem diameter at 0.70 mm. The single plant weight was recorded at 0.081 g, leaf area at 0.64 cm<sup>2</sup>, yield at 0.64 g/cm<sup>2</sup>, and dry matter ratio at 6.25%. The SPAD-chlorophyll value was 42.06. In T2, plant height increased to 7.62 cm, while stem diameter decreased to 0.64 mm. The single plant weight decreased to 0.069 g, and leaf area slightly reduced to 0.62 cm<sup>2</sup>; however, yield increased to 1.00 g/cm<sup>2</sup>. The chlorophyll content also showed a slight increase to 45.40. In T3, plant height further increased to 7.81 cm, maintaining the same stem diameter (0.70 mm), while the single plant weight decreased to 0.052 g. Leaf area was

reduced to 0.59 cm<sup>2</sup>, yet yield reached its maximum at 1.20 g/cm<sup>2</sup>, with a chlorophyll value of 43.08. Overall, despite higher sowing densities increasing yield, plant weight decreased, indicating increased competition for resources, which led to thinner stems. The chlorophyll content was highest at T2 density, while T1 exhibited the lowest value (Figure 1).



Figure 1. Image of broccoli microgreens subjected to different seed sowing density treatments Şekil 1. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan brokoli mikrofilizlerinin görüntüsü

## **Black Radish**

In black radish microgreens, under Full Surface Coverage Sowing (T1) conditions, plant height was measured at 12.60 cm, hypocotyl length at 8.91 cm, and stem diameter at 1.09 mm. The single plant weight was recorded at 0.26 g, leaf area at 1.41 cm<sup>2</sup>, yield at 0.65 g/cm<sup>2</sup>, and SPAD-chlorophyll value at 46.17. In T2 seed sowing density, plant height decreased to 10.89 cm, and stem diameter reduced to 1.01 mm. The single plant weight dropped to 0.186 g, while leaf area was measured at 1.15 cm<sup>2</sup>. However, the chlorophyll value increased to 49.67. At T3 seed sowing density, plant height further declined to 10.74 cm, with stem diameter recorded at 1.05 mm and single plant weight measured at 0.178 g. Leaf area and yield slightly decreased, but the SPAD-chlorophyll value peaked at 50.76. Overall, T1 resulted in the highest plant height and weight, while T3 seed sowing density provided the highest chlorophyll content, indicating an increase in chlorophyll concentration with higher sowing densities (Figure 2).



Figure 2. Image of black radish microgreens subjected to different seed sowing density treatments Şekil 2. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan siyah turp mikrofilizlerinin görüntüsü

## Red Beet

In red beet microgreens, the Full Coverage Sowing (T1) application resulted in a plant height of 7.23 cm, a hypocotyl length of 5.17 cm, and a stem diameter of 0.74 mm. The single plant weight was measured at 0.058 g, leaf area at 0.36 cm<sup>2</sup>, yield at 0.17 g/cm<sup>2</sup>, and the chlorophyll content was recorded at 31.96. Under T2 seed sowing density, plant height increased to 7.86 cm, stem diameter to 0.77 mm, and single plant weight to 0.061 g. The leaf area expanded to 0.43 cm<sup>2</sup>, the yield reached 0.23 g/cm<sup>2</sup>, and chlorophyll content improved to 33.67. At T3 seed sowing density, plant height slightly decreased to 7.81 cm, with a stem diameter of 0.78 mm, leaf area of 0.41 cm<sup>2</sup>, and chlorophyll content of 33.34. The results indicate that the T2 seed sowing density provided the best growth in

terms of plant height, leaf area, and yield, while chlorophyll content also remained highest at this density (Figure 3).



Figure 3. Image of red beet microgreens subjected to different seed sowing density treatments Şekil 3. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan kırmızı pancar mikrofilizlerinin görüntüsü

## Pea

In pea microgreens, the T1 application resulted in a plant height of 14.41 cm, a hypocotyl length of 7.10 cm, and a stem diameter of 1.76 mm. The single plant weight was recorded at 0.416 g, leaf area at 0.71 cm<sup>2</sup>, and chlorophyll content at 33.24. Under T2 seed sowing density, plant height increased to 14.51 cm, stem diameter to 1.82 mm, and single plant weight to 0.433 g. Leaf area expanded to 0.85 cm<sup>2</sup>, while the chlorophyll value decreased to 31.1. At T3 density, plant height slightly decreased to 14.29 cm; stem diameter was measured at 1.71 mm, and leaf area was reduced compared to T2 density. However, the chlorophyll value increased to 33.71. The data suggest that while T2 density promoted higher plant weight and leaf area, T3 density resulted in the highest chlorophyll content (Figure 4).



Figure 4. Image of pea microgreens subjected to different seed sowing density treatments Şekil 4. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan bezelye mikrofilizlerinin görüntüsü

## Sunflower

In sunflower microgreens, under T1 conditions, the plant height was measured at 12.08 cm, with a hypocotyl length of 8.40 cm and a stem diameter of 2.00 mm. The single plant weight was recorded at 0.884 g, leaf area at 3.39 cm<sup>2</sup>, yield at 0.67 g/cm<sup>2</sup>, and the chlorophyll value at 74.68. Under T2 seed sowing density, the plant height slightly decreased to 11.71 cm, while the stem diameter was measured at 1.91 mm, and the single plant weight declined to 0.821 g. Leaf area reduced to 2.93 cm<sup>2</sup>; however, yield increased to 0.81 g/cm<sup>2</sup>, with a chlorophyll value rising to 77.68. At T3 seed sowing density, the plant height was recorded at 11.65 cm, with a stem diameter of 1.95 mm. Leaf area decreased to 2.59 cm<sup>2</sup>, but yield increased to 0.92 g/cm<sup>2</sup>, and the chlorophyll value reached 81.62. In sunflower microgreens, higher sowing densities enhanced chlorophyll content, leaf area, and yield, while plant weight was greater at lower densities (Figure 5).



Figure 5. Image of sunflower microgreens subjected to different seed sowing density treatments Şekil 5. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan ayçiçek mikrofilizlerinin görüntüsü

## Bean

In bean microgreens, the T1 application resulted in a plant height of 24.16 cm, a hypocotyl length of 20.56 cm, and a stem diameter of 2.86 mm. The single plant weight was recorded at 2.910 g, and the chlorophyll value was 35.14. Under T2 density, the plant height slightly decreased to 23.57 cm, with a stem diameter of 2.71 mm, and the single plant weight dropped to 2.790 g. The chlorophyll value remained nearly constant at 35.03. At T3 density, the plant height further decreased to 23.26 cm, the stem diameter was recorded at 2.50 mm, and the single plant weight was 2.560 g. However, the chlorophyll value increased to 36.27. Overall, higher densities in bean microgreens resulted in a slight decrease in plant height and weight, while the chlorophyll value reached its highest level at T3 density.



Figure 6. Image of bean microgreens subjected to different seed sowing density treatments *Sekil 6. Farklı tohum ekim yoğunluğu uygulamalarına maruz kalan fasulye mikrofilizlerinin görüntüsü* 

Table 2. Significance levels in a three-way ANOVA analyzing the effects of plant type, seed sowing density, and their interactions

Çizelge 2. Bitki türü, tohum ekim yoğunluğu ve bunların etkileşimlerinin etkilerini analiz eden üç yönlü ANOVA'daki anlamlılık seviyeleri

	Plant species (Ps)	Seed Sowing Density (Ssd)	$Ps \times Ssd$	
Plant length	****	*	****	
Hypocotyl length	****	*	***	
Stem diameter	****	****	****	
Single plant weight	****	****	****	
Leaf Area	****	****	****	
SPAD-Chlorophyll	****	***	**	
Yield	****	****	****	
Dry Matter Ratio	****	**	****	

\*: p > 0.05, \*\*:  $p \le 0.05$ , \*\*\*:  $p \le 0.01$ , \*\*\*\*:  $p \le 0.001$ , ns: not significant.

Table 3. The effects of different seed sowing quantity treatments on plant height, hypocotyl length, stem diameter, single plant weight, leaf area, yield, dry matter, and SPAD-chlorophyll in microgreens

*Çizelge 3. Farklı tohum ekim miktarı uygulamalarının mikrofilizlerde bitki boyu, hipokotil uzunluğu, sap çapı, tek bitki ağırlığı, yaprak alanı, verim, kuru madde ve SPAD-klorofil üzerindeki etkileri* 

Factor		Plant length(cm)	Hypocotyl length (cm)	Stem diameter(mm)	Single plant weight (g)	Leaf A (cm²/plant)	Area	Yield (g/cm²)	Dry Matter Ratio (%)	SPAD- Chlorophyll
Plant speci	es	0	• •	• •	0 0				• •	1 0
Broccoli		7.41 e	5.70 d	$0.68~{ m f}$	0.067 e	0.62 e		0.95 a	5.78 d	43.51 с
Black radis	sh	11.41 d	8.63 b	1.05 d	0.208 d	1.20 c		0.89 b	5.09 e	48.87 b
Red beet		7.63 e	5.41 d	0.76 e	0.060 e	$0.40 \mathrm{f}$		0.23 e	4.16 f	32.99 e
Pea		14.41 b	7.19 с	1.76 с	0.420 c	0.78 d		0.21 e	9.54 b	32.95 e
Sunflower		11.51 с	8.38 b	1.95 b	0.840 b	$2.97 \mathrm{b}$		0.80 c	7.88 с	77.99 a
Bean		23.66 a	20.03 a	2.69 a	2.753 a	15.11 a		0.71 d	11.45 a	35.48 d
P value		≤,0001*	≤,0001*	≤,0001*	≤,0001*	≤,0001*		≤,0001*	≤,0001*	≤,0001*
LSD		0.3872	0.3415	0.0427	0.0181	0.0497		4.2032	0.2850	1.8074
Seed sowin	g den	sity								
T1	-	12.88 a	9.19 a	1.52 a	0.76 a	3.81 a		$0.50 \mathrm{c}$	7.18 b	43.88 b
T2		12.69 ab	9.13 a	1.47 b	0.72 b	3.34 c		0.67 b	7.30 ab	45.56 a
Т3		$12.59 \mathrm{b}$	9.35 a	1.45 b	0.67 c	3.38 b		0.72 a	7.48 a	46.46 a
P value		0,1146	0,1837	≤,0001*	≤,0001*	≤,0001*		≤,0001*	0,0189*	0,0006*
LSD		0.2738	0.2415	0.0302	0.0128	0.0351		2.9721	0.2015	1.2780
Plant speci	es×Se	ed sowing dens	sity.							
Broccoli	T1		5.03 h	0.70 kl	0.081 1	0.64 lm		$0.64~{ m f}$	$6.25~\mathrm{f}$	42.06 e
	T2	$7.62~{ m g}$	$5.89~{ m fg}$	0.641	0.069 1	0.62 m		1.00 b	$5.68~{ m g}$	45.40 d
	T3	7.81 g	$6.18 \mathrm{f}$	0.70 kl	0.052 1	0.59 m		1.20 a	5.41 gh	43.08 de
Black radis	hT1	12.60 d	8.91 c	1.09 h	$0.26~{ m g}$	$1.41~{ m g}$		$0.65~{ m f}$	4.56.1	46.17 d
	T2	$10.89 { m f}$	8.45 c	1.01 1	0.186 h	1.15 h		1.00 b	5.07 h	49.67 с
	T3	$10.74 \mathrm{~f}$	$8.52~\mathrm{c}$	1.05 hı	0.178 h	1.05 1		1.03 b	$5.63~{ m g}$	50.76 с
Red beet	T1	$7.23~{ m gh}$	5.17 h	0.74 jk	0.058 1	0.36 n		0.17 j	4.17 ıj	$31.96 \mathrm{~g}$
	T2	$7.86~{ m g}$	$5.53~{ m gh}$	0.77 jk	0.061 1	0.43 n		0.23 h	4.04 j	$33.67 \mathrm{~fg}$
	T3	$7.81~{ m g}$	$5.54~{ m gh}$	0.78 j	0.061 1	0.41 n		$0.30 \mathrm{~g}$	4.28 ıj	33.34  fg
Pea	T1	14.41 с	7.10 de	$1.76~{ m fg}$	$0.416~{ m f}$	0.71 kl		0.18 ıj	9.66 c	$33.24~\mathrm{fg}$
	T2	14.51 c	6.86 e	1.82 f	$0.433~{ m f}$	0.85 j		0.22 h	9.34 c	31.1 g
	T3	14.29 с	7.61 d	$1.71~{ m g}$	$0.411~{ m f}$	0.78 jk		0.24 h	9.63 c	33.71  fg
Sunflower	T1	12.08 de	8.40 c	2.00 d	0.884 d	3.39 d		$0.67 { m ef}$	7.55 e	74.68 b
	T2	11.71 e	8.36 c	1.91 e	0.821 e	2.93 e		0.81 d	7.91 de	$77.68 \mathrm{b}$
	T3	11.65 e	8.38 c	1.95 de	0.815 e	$2.59~{ m f}$		0.92 c	8.19 d	81.62 a
Bean	T1	24.16 a	20.56 a	2.86 a	2.910 a	16.38 a		0.70 e	10.92 b	$35.14~\mathrm{f}$
	T2	23.57 ab	19.68 b	2.71 b	2.790 b	14.06 c		0.78 d	11.74 a	$35.03~{ m fg}$
	T3	23.26 b	19.86 b	2.50 с	2.560 с	14.91 b		0.92 c	11.71 a	$36.27~\mathrm{f}$
P value		≤,0001*	0,0016	≤,0001*	≤,0001*	≤,0001*		≤,0001*	≤,0001*	0,0436*
LSD		0.6706	0.5916	0.0740	0.0314	0.0861		7.2802	0.4936	3.1306

T1: Seeds are sown to fully cover the surface area of the containers. T2: Seeds are sown at 50% above the surface area capacity, T3: Seeds are sown at 100% above the surface area capacity. *\*There are no statistical (p (0.05) differences between values with the same letters in the same columns.* 

## DISCUSSION

Although it is well known that different seed sowing density applications significantly affect yield outcomes, these effects vary depending on the species. Dubey et al. (2024) emphasized the importance of seed sowing density in the microgreens literature due to its direct impact on the growth of microgreens. In this study, it was observed that increasing seed sowing density led to a rise in the overall yield of microgreens. However, a proportional decrease in single plant weight was noted in broccoli, black radish, sunflower, and bean microgreens. In contrast, in red beet and pea microgreens, a reduction in single plant weight was observed only beyond the optimal growth level (Table 3). Choe et al. (2018) reported a linear relationship between seed sowing density and fresh weight yield. However, the findings of this study, while supporting this relationship, also demonstrate that high seed sowing density leads to a reduction in single plant weight. This outcome indicates increased resource competition among plants, where limited space and nutrient availability adversely affect the growth conditions of individual seedlings. Microgreens grown at high densities are subjected to reduced access to light, water, and nutrients per plant, resulting in a significant decline in individual development. Lee et al. (2004) reported a linear relationship between seed sowing density and yield in beet and chard microgreens. Similarly, Murphy et al. (2010) observed a comparable relationship between seed sowing density and yield in arugula (rocket). As seed sowing density increases, yield also improves; however, the associated seed costs must be carefully evaluated. Sariver et al. (2024) noted that densely sown okra, garden cress, and spinach microgreens exhibited higher yield values compared to sparsely sown counterparts. Consistent with these findings, this study also observed an increase in yield with higher seed density. When evaluating yield performance and the amount of seeds used, it is crucial to consider seed input to ensure an accurate yield performance analysis. A similar study conducted by Cowden et al. (2024) highlighted that the effects of increased density on biomass exhibited a similarly non-linear relationship, emphasizing that such changes in density do not always result in the expected yield improvements.

In broccoli microgreens, an increase in seed sowing density was observed to result in taller plants, while in red beet and pea microgreens, plant height increased until reaching the optimal growth level (Table 3). This phenomenon can be attributed to the closer spacing of microgreens at higher seed sowing densities, leading to competition for sunlight (Ntsoane et al., 2023). The height of microgreens plays a significant role in facilitating manual harvesting. Research indicates that taller microgreens are generally easier to harvest, as increased height simplifies the process (Palmitessa et al., 2020). Senevirathne et al. (2019) recommend an optimal harvest height of approximately 6 cm for maximum efficiency. A study conducted by Lerner et al. (2024) observed that microgreens exceeding this height exhibited a tendency to bend due to excessive elongation of the hypocotyls. This was reported as an undesirable trait from a commercial perspective, as it negatively impacts both visual appeal and post-harvest processing. In this study, we found that the height and hypocotyl length of microgreens varied depending on the species. Notably, as the height and hypocotyl length of large-seeded microgreens increased, bending and curling were observed in bean and pea microgreens. Priti et al. (2022) stated that in microgreens, seed density is directly related to seed size. This result aligns with findings by Panyapruek et al. (2016), who reported that lettuce grown at low plant density developed thicker stems. Differences in species' responses to seed sowing density likely reflect their genetic traits, physiological processes, and adaptive mechanisms to environmental factors. In densely growing species such as broccoli, this can lead to increased light competition, resulting in elongated but thinner stems. To better understand the underlying causes of such differences, in-depth investigations into genetic composition, physiological responses, and species-specific resource utilization strategies are necessary.

Overall, it was observed that an increase in seed sowing density led to a reduction or partial reduction in stem diameter across all microgreens species except for red beet (Table 3). This variation may be attributed to the stronger adaptability of red beet to high seed sowing density. Red beet appears to be a species that thrives better under dense conditions and is more resistant to competition, which may prevent a reduction in stem diameter. In contrast, for other microgreens species, high seed sowing density likely led to increased competition among plants, making access to nutrients and water more challenging and resulting in reduced stem diameter. The differing response of red beet to this condition could be explained by a combination of genetic and environmental factors. In the study by Balik et al. (2024), it was noted that certain environments were more effective in terms of plant height, hypocotyl development, stem diameter, and yield, while other environments performed less effectively. The study demonstrated that some environments yielded moderate results, whereas others were less effective in promoting plant growth due to factors such as low water retention capacity, insufficient nutrient support, and the inability to provide suitable structures for root development. The findings align with these observations, suggesting that without selecting an appropriate growing medium, seeds may encounter developmental challenges. This is further supported by the results of Ntsoane et al. (2023), which show a reduction in stem diameter with increasing seed sowing density in radish, cabbage, and arugula microgreens.

In this study, we observed that as seed sowing density increased, the leaf area of microgreens decreased in some species, while in others, it showed a partial decrease. This finding is consistent with the results of Maboko et al.

(2009), who reported that high seed sowing density in microgreens can increase the incidence of fungal diseases, thereby reducing the quality of the microgreens. Similarly, Signore et al. (2024) observed that rapini seedlings grown at the lowest seed sowing density exhibited more developed and larger true leaves compared to those grown at the highest density.

Chlorophyll pigment is essential for plants to carry out photosynthesis and significantly influences growth and yield (Hasanuzzaman & Fujita, 2022). Chlorophyll content generally increases with higher seed sowing density, aligning with the findings of Ntsoane et al. (2023). The potential contribution of increased chlorophyll content at higher seed sowing densities to photosynthetic capacity and product quality warrants further investigation. While increased chlorophyll typically supports photosynthetic efficiency, this effect may vary depending on the growing environment, genetic traits, and environmental factors. The observation of lower chlorophyll levels in some species under low seed density may indicate species-specific responses and adaptation mechanisms. To better understand these differences, more specific and controlled studies are needed.

In this study, a decrease in dry matter content was observed in broccoli microgreens, while in other species, it either increased or showed a partial increase. Several potential reasons could explain the reduction in dry matter content in broccoli microgreens. Although broccoli exhibited higher growth and yield compared to other species, this may have led to an increase in water content, consequently lowering the dry matter ratio. In microgreens, increases in yield and size are typically accompanied by a rise in water content, which suggests that while the total biomass of the plant may increase, the dry matter percentage could decrease. Additionally, the fast growth rate of broccoli may have accelerated photosynthesis and carbon accumulation, leading to higher water retention, which could negatively affect dry matter content. Changes in seed sowing density also influence dry matter content in microgreens; higher dry matter content generally extends the shelf life of microgreens (Sánchez et al., 2018; Valverde-Miranda et al., 2021). Producers should be informed not only about the variables examined in this study (growth stage, harvest height and leaf characteristics, yields) but also about additional factors such as fungal infections when selecting sowing densities (Nolan, 2018).

## CONCLUSION

This study observed that as seed sowing density increased, the results varied according to the genetic differences of the species. From an economic perspective, an increase in seed sowing density in broccoli microgreens resulted in higher yields and better quality products. This suggests that producers may consider increasing sowing density during broccoli microgreens production, as denser sowing can lead to more plants and thus higher yields. However, in red beet and pea microgreens, partial positive results were observed after reaching the optimal growth level; this emphasizes the importance of determining the ideal seed sowing density for each species.

Economically, determining the appropriate seed sowing density can optimize production costs while also improving productivity and quality. Excessive seed density can increase competition between plants for nutrients and water, which may lead to lower yields. Therefore, producers, particularly for fast-growing species like broccoli, need to carefully determine the sowing density, taking into account the economic benefits of higher sowing density. These findings can help microgreens producers develop more efficient and cost-effective production strategies. However, further research is needed, particularly to conduct more in-depth analyses of the economic impact of seed sowing density on different species.

## ACKNOWLEDGMENTS

## Contribution Rate Statement Summary of Researchers

The authors declare that they have contributed equally to the article.

## Conflict of Interest

The authors declare that there is no conflict of interest between them.

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