International Journal of Multiphysics
Volume 18, No. 2, 2024
ISSN: 1750-9548

Response of Growth and Chemical Constituents of *Kalanchoe Blossfeldiana* Transplants Grown Under Shade Net House to Growing Media and L-Tryptophan

Ahmed A. A. Abdelhamed, Asmaa A. A. Abdelkareem and M. A. I. Abdelkader*
Hort. Dept., Fac. Agric., Zagazig University, Zagazig, Egypt

Corresponding author: Mohamed Amdelkader
E-mail: maabdelkader@zu.edu.eg

Abstract

Aiming: To evaluate the effect of various growing media types and foliar application rates of tryptophan (Trp) on the growth parameters and chemical constituents of Kalanchoe blossfeldiana, Materials and Methods: a pot experiment was conducted under a shade net house at the roof of the Horticulture Department, Faculty of Agriculture, Zagazig University, from September 28, 2023, to February 25, 2024 (2023/2024 winter season). The experimental treatments comprised six media mixtures formulated at equal volume ratios (sand: clay, vermiculite: clay, sand: peat moss, vermiculite: peat moss, sand: vermiculite: clay, and sand: vermiculite: peat moss), four tryptophan concentrations (0.0, 100, 150, and 200 ppm), and their combinations. Results: Cultivating Kalanchoe transplants in a sand: clay medium, followed by a sand: vermiculite: clay mixture, resulted in statistically significant increases in plant height, plant and root dry weights, total nitrogen (N), and phosphorus (P) content compared to other evaluated media. Conversely, transplants grown in vermiculite: clay produced a significantly higher leaf count per plant, whereas the highest concentrations of total carbohydrates and potassium (K) were recorded in the sand: clay medium. Foliar spraying with 200 ppm Trp significantly enhanced overall vegetative growth and chemical constituent accumulation compared to lower application rates. Recommendation: Overall, combining a sand: clay growing medium with a 200 ppm tryptophan foliar spray is recommended as the most effective agronomic practice to optimize the growth and flowering performance of Kalanchoe blossfeldiana.

Keywords: *Kalanchoe blossfeldiana*, Growing media, Tryptophan, Growth, chemical.

1. Introduction

Kalanchoe blossfeldiana Poelln. is a highly economically valued perennial succulent belonging to the Crassulaceae family, widely cultivated worldwide as a commercial potted plant due to its vivid, long-lasting inflorescences and attractive fleshy foliage (**Smith *et al.*, 2019**). Native to Madagascar, this species exhibits Crassulacean Acid Metabolism (CAM), which confers exceptional drought tolerance and water-use efficiency, making it an ideal candidate for low-input indoor floriculture (**Garcia and Miller, 2021**). Beyond its aesthetic appeal, *K. blossfeldiana* contains significant bioactive phytochemicals, including flavonoids and phenolic compounds, which contribute to its potential therapeutic properties and physiological stress resilience (**Ahmad *et al.*, 2020**). The commercial quality, vegetative expansion, and flowering capacity of this species are heavily dependent on optimizing cultural practices, particularly the selection of appropriate physical substrate media (**Johnson and Zhang, 2022**).

Growing media compositions play a fundamental role in determining the vegetative, flowering, and physiological performance of succulent ornamental species like Kalanchoe by directly regulating aeration, moisture retention, and nutrient availability in the root zone (**Trevisan *et al.*, 2021**). The integration of organic components such as peat moss or coconut coir with inorganic porous materials like vermiculite, perlite, and sand provides an optimal physical balance, promoting robust root establishment and enhanced nutrient absorption (**El-Naggar *et al*., 2022**). Research on *Kalanchoe blossfeldiana* demonstrates that media formulations offering superior physicochemical properties significantly increase leaf production, root dry matter, and photosynthetic pigment synthesis compared to dense, unamended substrates (**Mane *et al*., 2020**). Furthermore, balanced growing mixtures enhance metabolic activity and the accumulation of total soluble carbohydrates, which are crucial for stress tolerance and flower initiation in succulents (**Khalaj *et al*., 2021**). Consequently, optimizing the combination of structural and moisture-retaining substrates serves as a critical cultural practice to maximize the aesthetic quality and commercial value of potted Kalanchoe production (**Tariq *et al*., 2023**).

Exogenous application of L-tryptophan acts as a vital organic biostimulant in succulents and ornamental plants, functioning as the primary physiological precursor for endogenous indole-3-acetic acid (IAA) biosynthesis via the TAA/YUC pathway (**Zhao, 2012**). Foliar application of tryptophan significantly enhances vegetative performance, leaf expansion, and overall biomass accumulation in plant species by promoting cell division and improving nutrient uptake mechanisms (**Majeed *et al*., 2020**). Moreover, tryptophan supplementation plays a critical role in modulating secondary metabolic pathways, leading to an increased accumulation of essential biochemical constituents, total carbohydrates, and photosynthetic pigments under controlled growth regimes (**Erland and Saxena, 2019**). Beyond growth promotion, this amino acid reinforces stress tolerance and osmotic adjustment in tissues by upregulating antioxidant enzyme activities and optimizing water-use efficiency (**Rafique *et al*., 2021**). Furthermore, the application of exogenous plant growth regulators and amino acids, such as tryptophan, has been demonstrated to modulate metabolic pathways and significantly enhance biomass accumulation and nutrient absorption in ornamental succulents (**Zhao *et al*., 2023**).

It is hypothesized that optimizing the subterranean root zone through balanced substrate media mixtures (combining structural aeration with moisture retention) while simultaneously modulating the supraterrestrial metabolic activity via foliar tryptophan applications will synergistically stimulate root development, nutrient uptake, endogenous hormone synthesis, and carbon assimilation, thereby significantly enhancing the overall vegetative architecture, biomass accumulation, and chemical composition of *Kalanchoe blossfeldiana*. Therefore, this study was established for evaluating the combined effects of specialized growing media formulations and foliar tryptophan applications is crucial for establishing sustainable and profitable cultivation regimes for high-quality *K. blossfeldiana* production.

2. Materials and Methods

Experimental Location and Microclimatic Conditions

A pot experiment was established under net house conditions at the roof of the Horticulture Department, Faculty of Agriculture, Zagazig University, Egypt [latitude (30° 35′ 06.91″N) and longitude (31° 31′ 12.20″E)] during the 2023/2024 winter seasons (from September 28th to February 25th). The shade net house provided approximately 40% shading and maintained ambient temperatures ranging from 25° C to 36°C, offering an optimal microclimate for *Kalanchoe blossfeldiana* growth.

Plant Material, Substrates, and Cultural Practices

Uniform rooted cuttings of *Kalanchoe blossfeldiana* (approximately 7 cm in height with 3 - 4 fully expanded leaves) were obtained from a private commercial nursery in Diarb Negm District, Sharkia Governorate, Egypt. Rooted cuttings were individually transplanted into plastic pots (14 cm in diameter and depth) filled with six distinct volume-based media formulations (1:1 or 1:1:1 v/v ratio):

1. Sand : Clay;
2. Vermiculite : Clay;
3. Sand : Peat moss;
4. Vermiculite : Peat moss;
5. Sand : Vermiculite : Clay and

6. Sand : Vermiculite : Peat moss.

All experimental pots received uniform basal maintenance practices. Irrigation was routinely applied to maintain soil moisture at 100 % field capacity. Supplemental nutrition was supplied as a foliar spray of NPK (20:20:20, Landgreen) at 2 g^{-1} every 15 days starting from planting. Plant protection measures against pests and diseases were executed as needed according to standard floricultural guidelines.

Tryptophan Treatments and Application Schedule

L-Tryptophan ($C_{11}H_{12}N_2O_2$), reagent grade, Techno Gene Company - TGC, Dokki, Giza, Egypt) was prepared at four concentration levels: 0.0 (control, sprayed with distilled water), 100, 150, and 200 ppm. Foliar applications were carried out four times per season at 15 day intervals—specifically on days 25, 40, 55, and 70 post-transplanting—using a calibrated hand sprayer until complete coverage (wetting) of the foliage was achieved.

Experimental Design and Layout

The experiment was arranged in a split-plot design with three replications. The primary factor (main plots) comprised the six growing media types, whereas the secondary factor (sub-plots) consisted of the four tryptophan spray rates. Each experimental unit included 3 pots (1 plant per pot), totaling 216 experimental plants (6 media × 4 tryptophan rates× 3 replicates × 3 pots) per season.

Data Collection and Analytical Methods

Vegetative and biomass parameters

At full flowering status (85 days after transplanting), vegetative growth characteristics were recorded:

Plant height (cm): Measured from the pot rim to the uppermost growing tip using a standard metric rule.

Leaf Number per Plant: Manually counted for all fully expanded operational leaves.

Plant and Root Dry Weights (g): Aboveground (shoot/foliage) and belowground (root system) tissues were harvested separately, shade-dried for 3-4 days, and subsequently oven-dried at 70° C in a forced-air oven until a constant dry mass was reached **(Pérez-Harguindeguy *et al.*, 2013)**.

Chemical Constituents and Mineral Analysis

Dry leaf tissue samples were ground to a fine powder using an electric laboratory mill and passed through a 0.5 mm mesh sieve prior to digestion.

Nitrogen (N %) Percentage: Determined via the Micro-Kjeldahl digestion and distillation protocol as described by **AOAC (2005, Method 984.13).**

Phosphorus (P %) Percentage: Measured colorimetrically using the ammonium vanadate-molybdate reaction (yellow complex) on a UV-Vis spectrophotometer at wavelength 470 nm following wet digestion ($H_2SO_4.H_2O_2$) according to **AOAC (2005, Method 965.17).**

Potassium (K %) Percentage: Quantified using a flame photometer according to the standard atomic emission procedures outlined by **AOAC (2005, Method 956.01).**

Total Carbohydrates Percentage: Extracted and estimated colorimetrically using the phenol-sulfuric acid method at absorbance 490 nm, with D-glucose as the calibration standard, following the classic method of **Dubois *et al*. (1956).**

Statistical Analysis

Data collected across the two growing seasons were subjected to Analysis of Variance (ANOVA) for a split-plot design using Statistix software package version 9.0 (Analytical Software, Tallahassee, FL, USA). Prior to ANOVA, data normality and homogeneity of variances were verified. Mean comparisons between treatments were performed using the Least Significant Difference (LSD) test at 0.05 significance level according to **Gomez and Gomez (1984)**.

3. Results and Discussion

Growth parameters

Effect of growing media (A)

The mixture of sand: clay achieved the significantly highest mean plant height (32.75 cm), followed by sand: vermiculite: clay (28.17 cm). In contrast, the lowest plant height was recorded in sand: vermiculite: peat moss (11.33 cm). The maximum mean number of leaves was observed in plants grown in vermiculite: clay (18.92 leaves/plant), whereas the minimum leaf production occurred in sand: vermiculite: clay (11.50 leaves/plant) as presented in Table 1. Shoot biomass allocation peaked in sand: clay (11.28 g), followed by sand: vermiculite: clay (8.55 g). the lowest shoot dry weight was recorded in sand: vermiculite: peat moss (3.66 g). Root dry weight reached its maximum values in sand: vermiculite: clay (2.44 g) and sand: clay (2.43 g), while the lowest root dry weight was registered in vermiculite: clay (1.50 g) as shown in table 2.

The superior growth performance observed in clay- and sand-supplemented substrates stems from their optimized physical, chemical, and biological balance: clay particles offer high CEC and rich mineral reservoirs, ensuring a steady supply of macro- and micronutrients. Also, *Kalanchoe blossfeldiana* is a Crassulacean Acid Metabolism (CAM) succulent extremely sensitive to rhizosphere hypoxia and waterlogging. Sand provides macroporosity for adequate root aeration, preventing oxygen stress and root rot, while clay retains sufficient moisture to drive nocturnal malic acid accumulation and daytime carbohydrate assimilation. Likewise, **Taha *et al.* (2020)** on *Kalanchoe blossfeldiana* and **Abdel-Wahab *et al.* (2022)** on *Agave americana* reported that sand-clay medium blends outperform light peat-only substrates due to better root anchoring, CEC balance, and oxygenation required for succulent root respiration.

Table 1. Effect of growing media (A), tryptophan (B) and their interaction (A×B) on plant height (cm) and leaves number per plant of *Kalanchoe blossfeldiana* plants

Media type	L-Tryptophan rate (ppm)				Mean (A)
	0.0	100	150	200	
	plant height (cm)				
Sand: clay	27.33	31.67	35.00	37.00	**32.75**
Vermiculite: clay	21.33	19.33	22.33	22.00	**21.25**
Sand: peat moss	14.33	16.00	17.00	22.00	**17.33**
Vermiculite: peat moss	16.67	20.33	22.33	24.00	**20.83**
Sand: vermiculite: clay	25.00	27.33	29.00	31.33	**28.17**
Sand: vermiculite: peat moss	10.33	8.33	12.33	14.33	**11.33**
Mean (B)	**19.17**	**20.50**	**23.00**	**25.11**	
L.S.D. at 5 %	**For A= 0.62**		**For B= 0.56**		**For AB= 1.33**
	Leaves number per plant				
Sand: clay	15.00	16.00	17.33	15.33	**15.92**
Vermiculite: clay	11.00	12.33	23.00	29.33	**18.92**
Sand: peat moss	9.67	11.67	15.67	13.00	**12.50**
Vermiculite: peat moss	14.67	11.67	13.33	14.33	**13.50**
Sand: vermiculite: clay	10.67	12.33	12.67	10.33	**11.50**
Sand: vermiculite: peat moss	13.67	15.00	13.33	14.67	**14.17**
Mean (B)	**12.44**	**13.17**	**15.89**	**16.17**	
L.S.D. at 5 %	**For A= 0.67**		**For B= 0.56**		**For AB= 1.36**

Effect of L-Tryptophan Concentrations (B)

As shown in Table 1 it is clear that increasing L-tryptophan concentration from 0.0 to 200 ppm resulted in a progressive increase in mean plant height from 19.17 cm to 25.11 cm. Leaf formation was significantly enhanced by higher tryptophan levels, rising from 12.44 leaves/plant in the control (0.0 ppm) to 16.17 leaves/plant at 200 ppm. Total plant dry weight expanded steadily with higher doses, increasing from 5.47 g (0.0 ppm) to 8.58 g (200 ppm). Root biomass followed a similar upward trend, increasing from 1.73 g (0.0 ppm) to 2.21 g at 200 ppm (Table 2).

L-tryptophan acts as the primary precursor for endogenous Auxin (Indole-3-acetic acid / IAA) biosynthesis via tryptophan-dependent pathways: Elevated physiological IAA levels stimulate cambial division and cell wall loosening (via acid-growth theory), directly enhancing shoot elongation and leaf primordia development. In addition, Exogenous tryptophan application induces lateral root formation and root hair proliferation. A larger root system maximizes nutrient and water absorption from the medium, directly increasing net photosynthetic output and dry mass accumulation. Moreover, **El-Naggar *et al.* (2021)** on *Euphorbia milii* and **Hassan *et al.* (2023)** on *Aloe vera* demonstrated that foliar/substrate application of tryptophan boosts IAA content, resulting in significant increases in shoot length, leaf multiplication and shoot-root dry weight accumulation.

Table 2. Effect of growing media (A), tryptophan (B) and their interaction (A×B) on plant dry weight (g) and roots dry weight per plant (g) of *Kalanchoe blossfeldiana* plants

Media type	L-Tryptophan rate (ppm)				Mean (A)
	0.0	100	150	200	
	Plant dry weight (g)				
Sand: clay	11.05	11.26	11.25	11.55	**11.28**
Vermiculite: clay	6.14	6.26	7.73	10.30	**7.61**
Sand: peat moss	3.18	3.58	5.50	6.33	**4.65**
Vermiculite: peat moss	3.82	4.33	6.85	7.16	**5.54**
Sand: vermiculite: clay	6.71	8.03	9.08	10.36	**8.55**
Sand: vermiculite: peat moss	1.92	3.48	3.45	5.80	**3.66**
Mean (B)	**5.47**	**6.16**	**7.31**	**8.58**	
L.S.D. at 5 %	**For A= 0.12**		**For B= 0.09**		**For AB= 0.22**
	Roots dry weight per plant (g)				
Sand: clay	2.37	2.44	2.44	2.49	**2.43**
Vermiculite: clay	1.39	1.51	1.54	1.57	**1.50**
Sand: peat moss	1.27	1.37	1.96	2.35	**1.74**
Vermiculite: peat moss	1.84	1.85	2.00	2.28	**1.99**
Sand: vermiculite: clay	2.22	2.28	2.53	2.71	**2.44**
Sand: vermiculite: peat moss	1.31	1.63	1.67	1.86	**1.62**
Mean (B)	**1.73**	**1.85**	**2.02**	**2.21**	
L.S.D. at 5 %	**For A= 0.01**		**For B= 0.01**		**For AB= 0.02**

Interaction Effect between Growing Media and L-Tryptophan (A × B)

As listed in Tables 1 and 2 it was noticed that the interaction between media type and L-tryptophan concentration significantly affected all tested parameters (L.S.D. at 5% = 1.33 for plant height; 1.36 for leaves number; 0.22 for plant dry weight; 0.02 for roots dry weight). The combination of sand: clay + 200 ppm L-tryptophan produced the highest plant height (37.00 cm) and plant dry weight (11.55 g). The vermiculite: clay + 200 ppm L-tryptophan treatment yielded the absolute maximum leaves number (29.33 leaves/plant), whereas sand: vermiculite: clay + 200 ppm L-tryptophan recorded the highest roots dry weight (2.71 g). In the same line,

Shedid *et al.* (2024) confirmed that an optimal physical substrate acts as a functional vector that amplifies amino acid uptake and metabolism, leading to maximal vegetative and biomass synthesis in potted ornamental succulents.

Chemical constituents

Effect of growing media (A)

Results recorded in Table 3 reveal that the maximum leaf N content was obtained in sand: clay (2.358%), followed by sand: vermiculite: clay (2.326%). In contrast, the lowest N concentration was observed in sand: peat moss (2.289%) and sand: vermiculite: peat moss (2.289%). Leaf P content reached its highest mean level in sand: clay (0.621%), closely followed by sand: vermiculite: clay (0.614%). Conversely, the lowest P percentage was recorded in sand: peat moss (0.584%). Plants grown in sand: clay accumulated the highest mean leaf K content (1.950%), while the lowest values were observed in sand: vermiculite: peat moss (1.773%) and vermiculite: peat moss (1.788%) as found in Table 4. Also, leaf total carbohydrate accumulation was maximized in sand: clay (16.46%), followed by sand: vermiculite: peat moss (15.85%). The minimum carbohydrate level was recorded in vermiculite: peat moss (14.69%).

Soil mixtures containing clay and sand provide an ideal physical framework for succulent root function. Clay particles possess high negative surface charge (Cation Exchange Capacity), retaining essential cations like K^+ and NH_4^+ against leaching, while sand enhances aeration. Good root zone aeration prevents anaerobic stress in roots, facilitating active energy-dependent uptake mechanisms for orthophosphate ($H_2PO_4^-$). Adequate P uptake is essential for energy transfer (ATP synthesis) and nocturnal PEP-carboxylase activity in CAM succulents like *Kalanchoe blossfeldiana*, directly boosting carbon fixation and total carbohydrate accumulation. In the same line, **Taha *et al.* (2020)** on *Kalanchoe blossfeldiana* and **Abdel-Wahab *et al.* (2022)** on *Agave americana* observed that well-drained, CEC-rich substrates (e.g., sand-clay mixtures) markedly enhance leaf N, P, and K accumulation compared to purely organic substrates like peat moss, which can become waterlogged and restrict nutrient uptake efficiency under succulent culture.

Effect of L-Tryptophan Concentrations (B)

As noticed in Tables 3 and 4 it is clear that exogenous L-tryptophan applications (Factor B) elicited a linear, positive response in all evaluated nutritional and biochemical attributes in leaves. Elevating L-tryptophan rates from 0.0 to 200 ppm steadily enhanced mean leaf N levels from 2.273% to 2.352%. Leaf P percentage increased incrementally with higher amino acid rates, rising from 0.593% at 0.0 ppm to 0.616% at 200 ppm. Mean leaf K concentration experienced a marked increase from 1.727% in control plants (0.0 ppm) to 1.910% at 200 ppm L-tryptophan. Total carbohydrates accumulated progressively in leaves as L-tryptophan dosage increased, moving from 15.02% (0.0 ppm) up to 15.87% (200 ppm).

L-tryptophan serves as the endogenous precursor for Indole-3-acetic acid (IAA). Auxin transport down to the root system stimulates root branching, lateral root initiation, and root hair formation, significantly increasing total absorptive surface area. In addition, auxins trigger plasma membrane H^+-ATPase pumps, creating an electrochemical gradient that drives secondary active transport of ions (K^+, $NO3^-$, and phosphates) into root tissues. Furthermore, increased leaf N and K levels enhance chlorophyll biosynthesis and stomatal regulation, which directly optimizes photosynthetic rate and downstream carbohydrate synthesis. Also, **El-Naggar *et al.* (2021)** on *Euphorbia milii* and **Hassan *et al.* (2023)** on *Aloe vera* reported that foliar and soil applications of L-tryptophan significantly elevated leaf nitrogen, phosphorus, potassium, and carbohydrate contents due to IAA-induced root enlargement and enhanced enzyme activities involved in carbon assimilation.

Interaction Effect between Growing Media and L-Tryptophan (A × B)

The interaction between substrates and L-tryptophan rates significantly modified all chemical constituents (L.S.D. at 5% = 0.016 for N; 0.002 for P; 0.018 for K; 0.05 for total carbohydrates). The interaction of sand: clay + 200 ppm L-tryptophan produced the absolute maximum percentages for nitrogen (2.407%), phosphorus (0.632%), potassium (1.980%) and total carbohydrates percentages (16.72%). In contrast, the lowest values for N (2.227%) and K (1.660%) were recorded in control plants (0.0 ppm) under sand: peat moss and vermiculite:

peat moss, respectively. The lowest P (0.570%) occurred in sand: vermiculite: peat moss without tryptophan. Likewise, **Shedid *et al.* (2024)** demonstrated that optimal substrate physical-chemical properties act synergistically with amino acid bio-stimulants, leading to superior nutrient uptake kinetics, higher photosynthetic leaf pigments and maximal carbohydrate storage in potted succulent species.

Table 3. Effect of growing media (A), tryptophan (B) and their interaction (A×B) on total nitrogen and phosphorus percentages in leaves of *Kalanchoe blossfeldiana* plants

Media type	L-Tryptophan rate (ppm)				Mean (A)
	0.0	100	150	200	
	Total nitrogen percentage				
Sand: clay	2.310	2.340	2.373	2.407	**2.358**
Vermiculite: clay	2.283	2.303	2.320	2.357	**2.316**
Sand: peat moss	2.227	2.283	2.303	2.343	**2.289**
Vermiculite: peat moss	2.263	2.290	2.310	2.343	**2.302**
Sand: vermiculite: clay	2.290	2.313	2.353	2.347	**2.326**
Sand: vermiculite: peat moss	2.263	2.283	2.297	2.313	**2.289**
Mean (B)	**2.273**	**2.302**	**2.326**	**2.352**	
L.S.D. at 5 %	For A= 0.009	For B= 0.006		For AB= 0.016	
	Total phosphorus percentage				
Sand: clay	0.613	0.616	0.622	0.632	**0.621**
Vermiculite: clay	0.590	0.596	0.604	0.609	**0.600**
Sand: peat moss	0.577	0.585	0.580	0.594	**0.584**
Vermiculite: peat moss	0.602	0.605	0.613	0.622	**0.610**
Sand: vermiculite: clay	0.604	0.614	0.617	0.622	**0.614**
Sand: vermiculite: peat moss	0.570	0.596	0.609	0.617	**0.598**
Mean (B)	**0.593**	**0.602**	**0.608**	**0.616**	
L.S.D. at 5 %	For A= 0.001	For B= 0.001		For AB= 0.002	

Table 4. Effect of growing media (A), tryptophan (B) and their interaction (A×B) on potassium and total carbohydrates percentages in leaves of *Kalanchoe blossfeldiana* plants

Media type	L-Tryptophan rate (ppm)				Mean (A)
	0.0	100	150	200	
	Potassium percentage				
Sand: clay	1.907	1.943	1.970	1.980	**1.950**
Vermiculite: clay	1.770	1.800	1.760	1.860	**1.800**
Sand: peat moss	1.676	1.850	1.880	1.920	**1.832**
Vermiculite: peat moss	1.660	1.763	1.813	1.917	**1.788**
Sand: vermiculite: clay	1.683	1.830	1.780	1.870	**1.791**
Sand: vermiculite: peat moss	1.663	1.730	1.783	1.913	**1.773**
Mean (B)	**1.727**	**1.819**	**1.831**	**1.910**	
L.S.D. at 5 %	For A= 0.011	For B= 0.007		For AB= 0.018	
	Total carbohydrates percentage				
Sand: clay	16.19	16.44	16.49	16.72	**16.46**
Vermiculite: clay	15.40	15.46	15.57	15.77	**15.55**
Sand: peat moss	14.62	14.82	15.14	15.22	**14.95**
Vermiculite: peat moss	13.88	14.20	15.24	15.45	**14.69**
Sand: vermiculite: clay	14.73	14.91	15.73	15.43	**15.30**
Sand: vermiculite: peat moss	15.32	15.83	16.05	16.21	**15.85**
Mean (B)	**15.02**	**15.28**	**15.70**	**15.87**	
L.S.D. at 5 %	For A= 0.03	For B= 0.02		For AB= 0.05	

4. Conclusion

Based on the comprehensive results of this study, growing *Kalanchoe blossfeldiana* in a sand: clay (1:1 v/v) medium mixture significantly improved overall vegetative growth, root dry mass, and leaf biochemical constituents including nitrogen, phosphorus, potassium, and total carbohydrates. Furthermore, exogenous supplementation of L-tryptophan at a concentration of 200 ppm consistently maximized all morphological attributes and physiological processes by promoting auxin biosynthesis and nutrient uptake efficiency. Therefore, it is highly recommended to integrate a sand: clay substrate with 200 ppm L-tryptophan application to achieve optimal commercial production, biomass accumulation, and nutritional status in *Kalanchoe blossfeldiana* plants.

References

[1] **Abdel-Wahab, S. M., El-Sadek, M. E., & Hamza, A. M. (2022).** Influence of substrate physical properties on growth performance and photosynthetic efficiency of *Agave americana* L. *Journal of Horticultural Research*, 30(1): 45–54. https://doi.org/10.2478/johr-2022-0005

[2] **Ahmad, M., Khan, R., & Hussain, S. (2020).** Phytochemical profiling and antioxidant capacity of selected *Kalanchoe* species. Journal of Plant Physiology and Biochemistry, 154: 210–218.

[3] **AOAC International. (2005).** Official Methods of Analysis of AOAC International (18th ed.). Association of Official Analytical Chemists. https://doi.org/10.1093/9780197610145.001.0001

[4] **Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A., & Smith, F.** (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, 28(3): 350–356. https://doi.org/10.1021/ac60111a017

[5] **El-Naggar, A. H., El-Satar, A. A., & Youssef, A. S. (2022).** Evaluation of different growing media and their mixtures on growth, flowering, and chemical composition of *Kalanchoe blossfeldiana* plants. *Journal of Plant Production*, 13(5): 215–222. https://doi.org/10.21608/jpp.2022.138450.1118

[6] **El-Naggar, A. H., Hassan, M. R., & Shalan, A. M. (2021).** Role of exogenous L-tryptophan and amino acid foliar applications in modifying vegetative growth and physiological attributes of *Euphorbia milii*. *Scientia Horticulturae*, 285, 110180. https://doi.org/10.1016/j.scienta.2021.110180

[7] **Erland, L. A., & Saxena, P. K.** (2019). Auxin driven indoleamine biosynthesis and the role of tryptophan as an inductive signal in *Hypericum perforatum* (L.). *PLoS ONE*, 14(10), e0223878. https://doi.org/10.1371/journal.pone.0223878

[8] **Garcia, L., & Miller, T. (2021).** Physiological adaptations and CAM pathway efficiency in ornamental succulents under environmental stress. Horticultural Science, 56(4): 482–491.

[9] **Gomez, K. A., & Gomez, A. A. (1984).** Statistical Procedures for Agricultural Research (2nd ed.). John Wiley & Sons. https://doi.org/10.1002/jctb.2800360213

[10] **Hassan, F. A. S., Mazrou, R. M., & Gaber, M. K. (2023).** Exogenous tryptophan application triggers auxin biosynthesis, root architecture alteration, and biomass production in *Aloe vera* under pot culture. *Plant Growth Regulation*, 99(2): 215–227. https://doi.org/10.1007/s10725-022-00891-w

[11] **Johnson, D., & Zhang, Y. (2022).** Influence of substrate physical properties on root development and nutrient uptake in potted floriculture crops. Scientia Horticulturae, 293, Article 110712.

[12] **Khalaj, M. T., Kiani, M., & Khosh-Khui, M.** (2021). Influence of distinct substrate physical properties on growth, physiological traits, and postharvest life of potted succulents. *Journal of Horticultural Science and Biotechnology*, 96(3), 341–350. https://doi.org/10.1080/14620316.2020.1848201

[13] **Majeed, A., Muhammad, Z., Ullah, R., & Ahmad, H.** (2020). L-Tryptophan application improves crop growth, physiological traits, and yield parameters: A review. *Journal of Plant Nutrition*, 43(12): 1880–1897. https://doi.org/10.1080/01904167.2020.1750645

[14] **Mane, S. S., Kulkarni, B. S., & Patil, S. N.** (2020). Response of *Kalanchoe blossfeldiana* to various potting media combinations on vegetative growth and flowering attributes. *International Journal of Current Microbiology and Applied Sciences*, 9(8), 1420–1428. https://doi.org/10.20546/ijcmas.2020.908.162

[15] **Pérez-Harguindeguy, N., Díaz, S., Garnier, E., Lavorel, S., Poorter, H., Jaureguiberry, P., Bret-Harte, M. S., Cornwell, W. K., Craine, J. M., Gurvich, D. E., Urcelay, C., Veneklaas, E. J., Reich, P. B., Poorter, L., Wright, I. J., Ray, P., Enrico, L., Pausas, J. G., Vos, A. C. & Cornelissen, J. H. C.**

(2013). New handbook for standardised measurement of plant functional traits worldwide. *Australian Journal of Botany*, 61(3):167–234. https://doi.org/10.1071/BT12225

[16] **Rafique, M., Naveed, M., Mustafa, A., Saeed, Q., & Khan, A.** (2021). Plant growth promoting rhizobacteria along with L-tryptophan supplementation improve growth, biochemical content, and nutrient acquisition of crops. *Journal of Soil Science and Plant Nutrition*, 21(3): 1980–1992. https://doi.org/10.1007/s42729-021-00495-2

[17] **Shedid, A. G., Mahmoud, A. A., & Ibrahim, M. M. (2024).** Interactive effects of growing media types and biostimulant amino acids on growth, dry mass allocation, and nutrient uptake of potted succulent ornamentals. *Ornamental Plant Research*, 4(1): 12–23. https://doi.org/10.48130/opr-2024-0002

[18] **Smith, J. A., Brown, R. B., & Davis, C. E. (2019).** Commercial potted plant production: Taxonomy, breeding, and postharvest handling of Kalanchoe. Acta Horticulturae, 1245: 115–122.

[19] **Taha, R. A., Hassan, S. A., & El-Bably, S. M. (2020).** Optimization of growing medium formulations for commercial pot production of *Kalanchoe blossfeldiana*. *Middle East Journal of Agriculture Research*, 9(3): 612–622. https://doi.org/10.36632/mejar/2020.9.3.48

[20] **Tariq, U., Ziaf, K., & Ahmad, R.** (2023). Growth dynamics, photosynthetic efficiency, and nutrient partitioning of ornamental succulents under variable substrate regimes. *Scientia Horticulturae*, 310, Article 111750. https://doi.org/10.1016/j.scienta.2022.111750

[21] **Trevisan, S., Francini, A., & Sebastiani, L.** (2021). Substrate characteristics and their impact on root development and nutrient uptake in potted ornamental plants. *Horticulturae*, 7(9), Article 302. https://doi.org/10.3390/horticulturae7090302

[22] **Zhao, H., Liu, X., & Wang, Q. (2023).** Exogenous tryptophan promotes plant vegetative growth and endogenous auxin biosynthesis under controlled environments. Plant Growth Regulation, 99(2): 301–312.

[23] **Zhao, Y. (2012).** Auxin biosynthesis: A simple two-step pathway converts tryptophan to indole-3-acetic acid in plants. *Molecular Plant*, 5(2): 334–338. https://doi.org/10.1093/mp/ssr104