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Micropropagation and Acclimatization of *Monstera deliciosa* Liebm. 'Thai Constellation'

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1. Introduction

Monstera deliciosa Liebm., also known as ceriman (Trinidad) or piñanona (Mexico), is a climbing vine (growing to a height of 30 ft or more) that belongs to the family Araceae [1,2]. It has been spread around the world as an ornamental foliage plant that can be used indoors or outdoors [3]. *M. deliciosa* 'Thai Constellation' is a new variant of *M. deliciosa*, possessing variegated leaves and being relatively genetically stable. Unfortunately, the plant grows slowly, and cuttings can be a challenge to root, driving up the price. The rapid propagation of *M. deliciosa* by tissue culture technology not only is conducive to the maintenance of excellent characters but also provides a large number of plantlets for plant cultivation [4].

In vitro clonal propagation of plants is popularly called micropropagation because of the miniaturization of the process [5]. Temporary immersion system (TIS) is one of the most advanced tools for commercial micropropagation. It works by using a semi-automated bioreactor to immerse cells, tissues, or organs in a liquid culture medium for a period of time. Hyperhydricity can be remedied by TIS, which exposes explants to liquid media intermittently rather than continuously [6]. Two variants of this system have been developed and are currently on the market: the Recipient for Automated Temporary Immersion system (RITA[®]) and the twin-flasks system (BIT[®]) [7]. More recently, a new bioreactor system has been developed (SETIS[™], Vervit, Lochristi, Belgium), which allows for increased light irradiation, automation of cultivation systems, easy handling, and large culture medium capacity, and successful protocols have been reported for the micropropagation of banana and vanilla [8,9,10].

The term acclimatization is defined as the climatic adaptation of an organism, especially a plant, that has been moved to a new environment [11,12].

Acclimatization is an important step in micropropagation. Biostimulants are defined as materials that contain substance(s) and/or microorganisms, whose function when applied to plants or the rhizosphere is to stimulate natural processes to enhance/benefit nutrient uptake, nutrient efficiency, tolerance to abiotic stress, and/or crop quality, independent of its nutrient content [13,14]. Protein hydrolysates (PHs) are a category of plant biostimulants defined as mixtures of polypeptides, oligopeptides, and amino acids that are manufactured from protein sources using partial hydrolysis [15,16]. Plant-derived PHs are gaining greater acceptance by farmers due to their richness in bioactive compounds and their great efficacy in enhancing crop performance [17]. The use of protein hydrolysis products as stimulants to assist plants in acclimatization seems to be a viable option at present.

The goal of this study was to evaluate the efficiency of the SETIS[™] bioreactor for the micropropagation of *M. deliciosa* 'Thai Constellation' as compared to a conventional semi-solid culture system. The development of a new micropropagation system for *M. deliciosa* may contribute to its large-scale commercialization. In view of the absence of data regarding the acclimatization of *M. deliciosa* 'Thai Constellation', the present study also had the objective of evaluating the effects of IQ Forte as a biostimulant on the acclimatization of in vitro-derived plantlets of *M. deliciosa* 'Thai Constellation'.

2. Materials and Methods

2.1. Plant Material and Culture Establishment

Plants of *M. deliciosa* 'Thai Constellation' were obtained from an ornamental nursery in Homestead, Florida, and in vitro cultures were established in the micropropagation laboratory of the Environmental Horticulture Dept., University of Florida, in Gainesville, FL. The axillary buds were collected and washed in the sink with soap and water, surface sterilized using 2% sodium hypochlorite (NaClO) for 20 min, and then rinsed in dH₂O (distilled water). Subsequently, the outer leaf layer and tissue were removed. The buds again were put in a fresh solution of 2% sodium hypochlorite for another 20 min and then rinsed 3 times. Explants were initially propagated in agar-based medium.

In vitro shoots were multiplied through axillary shoot multiplication in semi-solid modified MS medium [18] including B5 vitamins supplemented with 30 g/L sucrose, 2.5 g/L gellan gum, 7.5 mg/L BAP (6-benzylaminopurine), and 0.5 mg/L NAA (naphthalene acetic acid). The medium pH was adjusted to 5.6–5.8 before autoclaving at 121 °C for 25 min at 1.2 kg cm⁻². About 40 mL of medium was dispensed into baby food jars. This allowed the development of multiple axillary shoots and roots.

After 60 days, prior to subculture, shoots formed directly from axillary buds showed some roots and a small callus formation at the base of the shoot. In the subculture process, a small number of roots were trimmed, and the shoots were divided into single shoots containing a portion of the callus.

2.2. Acclimatization and Biostimulant

In vitro-derived shoots generated from bioreactors were used for this study. The acclimatization of in vitro-derived plantlets was conducted in a greenhouse.

Shoots were separated into single shoots under aseptic conditions and transplanted into baby food jars containing rooting medium. The rooting medium was a modified MS medium supplemented with 30 g/L sucrose, 2.5 g/L gellan gum, and 0.5 mg/L NAA (naphthalene acetic acid). The medium pH was adjusted to 5.6–5.8 before autoclaving at 121 °C for 20 min at 15 lbs pressure. About 40 mL of medium was dispensed into baby food jars. Cultures were maintained under fluorescent lighting (General Electric fluorescent bulbs 59W) at 50 μmol m⁻² s⁻¹ (FL), and the photoperiod was 16/8 h (light/dark). The temperature of the culturing room was set to 25 °C.

After 60 days, the plantlets with roots were transplanted to the greenhouse.

IQ Forte (IQBiotech, Miami, FL, USA), which is a natural fertilizer based on oligopeptides and free amino acids obtained by enzymatic hydrolysis of proteins from seeds, was used as the biostimulants.

2.3. In Vitro Multiplication Using Bioreactors

Young shoots (4–6 cm) collected from in vitro seedlings were used as explants for shoot proliferation.

Shoots were grown in four different culture systems: (1) in baby food jars containing 40 mL semi-solid medium (2.5 g/L gellan gum) as a control, (2) in temporary immersion bioreactors (SETIS™, VERVIT, Zelzate, Belgium) with an immersion and aeration frequency of 1.5 h and immersion duration of 1 min (Treatment 1), (3) in temporary immersion bioreactors with an immersion and aeration frequency of 1.5 h and immersion duration of 2 min (Treatment 2), (4) in temporary immersion bioreactors with an immersion and aeration frequency of 3 h and immersion duration of 2 min (Treatment 3) (Figure 1).

For the control group, two trays were prepared, each with 30 baby food jars (Figure 1). The 60 baby food jars in the two trays had identical experimental treatments. Cultures were maintained under fluorescent lighting (General Electric fluorescent bulbs 59W) at $50 \mu\text{mol m}^{-2} \text{s}^{-1}$ (FL), and the photoperiod was 16/8 h (light/dark). The temperature of the culturing room was $25 \pm 2 \text{ }^\circ\text{C}$. At the end of the experiment (90 days), 30 baby food jars were taken out to collect the experimental data.

The treatments in TIS were comprised of 4 replicates per treatment, each replicate containing 10 explants. The cultures were maintained under controlled environment conditions with continuous light (Valoya's L-Series LED grow light, model L35-144, 35W, AP67 spectrum; 14% Blue, 16% Green, 53% Red, and 17% Far Red) and light intensity of $79 \mu\text{mol m}^{-2} \text{s}^{-1}$, temperature of $25 \pm 2 \text{ }^\circ\text{C}$, and photoperiod of 16/8 h (light/dark). After 90 days of culture establishment, the initial 10 explants turned into 10 clusters with multiple shoots. Data were provided by 7 randomly selected clusters out of the 10. Plant height (height of the tallest shoot), fresh weight, dry weight, chlorophyll content, and shoot multiplication rate were evaluated. To obtain the dry weight, the plant material was placed in an oven and dried at $80 \text{ }^\circ\text{C}$ for 48 h. For the shoot multiplication rate, shoots that were clearly differentiated were counted, the total number was calculated, and they were classified into small, medium, and large grades based on their length (small: $\leq 2 \text{ cm}$, medium: 2–4 cm, large: $> 4 \text{ cm}$). Due to the low survival rate of small shoots in the acclimatization process, the medium and large shoots were summed up and referred to as 'effective shoots'.

It is important to note that in addition to the differences in the type of media used and the method of cultivation, the light source used in the bioreactor system is different from that of a traditional semi-solid system. The traditional semi-solid systems use regular fluorescent lights, as they are the traditional systems used in our lab for many different cultures, whereas for the bioreactor, we use the new LED system, as recommended for optimal growth, propagation, and development.

2.4. Relative Chlorophyll Content Analysis

Fully expanded leaves were selected from 5 plants per treatment for chlorophyll analysis. Chlorophyll relative content was measured as SPAD values by placing the third leaf of each plantlet, counted from top downwards, in a portable SPAD-502 chlorophyll meter (SPAD-502, Minolta Co., Ltd., Osaka, Japan).

2.5. Statistical Analysis

A completely randomized experimental design was used for all experiments. Data were collected and submitted to analysis of variance (ANOVA) using the software R version 4.1.3 (Copyright © 2022 The R Foundation for Statistical Computing). Tukey's post hoc multiple comparison adjustment ($\alpha = 0.05$) was used for all pairwise comparisons of means.

2.6. Effect of Concentration of Biostimulant on Plantlet Growth

The experiment was completely randomized with 3 treatments (3 concentrations of biostimulant; ml IQ Forte/L water) and one control group: (1) Treatment 1: 2.0 mL/L, (2) Treatment 2: 3.0 mL/L, (3) Treatment 3: 4.0 mL/L, (4) Control: water without biostimulant. The concentration of biostimulant was determined based on the manufacturer's label recommendations. The water in all treatments came from the irrigation system of the greenhouse with a water-soluble fertilizer (Peter's 20-10-20 Peat Lite at 150 ppm). There were 20 repetitions for each treatment, where one repetition means one plant in a pot. At the end of the experiment, the following parameters were evaluated: (1) survival: % of plants that survived after transplant to greenhouse, (2) growth: shoot and root development (length, fresh and dry weight), and (3) chlorophyll relative content (SPAD).

The biostimulant IQ Forte was applied at two-week intervals during the course of the experiment (according to the instructions provided by the manufacturer), 50 mL per pot per time. Meanwhile, foliar phosphite fertilizer was applied every two weeks to ensure that there was no microbial infection. The amount applied

was 50 mL per pot of an aqueous solution containing 1.6% of the original solution. The interval between the application of these two products was one week.

After 100 days of the plants growing in the greenhouse, data were collected. Survival rates were recorded first. Other data were provided by 10 randomly selected plantlets out of plants that survived. Plant height (height of the tallest shoot), fresh weight, dry weight, and chlorophyll content were evaluated.

3. Results

3.1. Micropropagation Using TIS Bioreactors

Significant differences were observed among culture systems for plant height. The highest plant height occurred in treatment 1 (1 min, 1.5 h; 5.2 cm), followed by treatment 2 (2 min, 1.5 h; 4.2 cm) and treatment 3 (2 min, 3 h; 4.0 cm). The control group in semi-solid medium had a plant height of 4.1 cm. The plant height of treatment 1 was significantly higher than the height in the other groups (Figure 2).

For fresh weight, treatments 1 and 2 ($p < 0.001$) (17.02 g and 18.13 g, respectively) were significantly higher than treatment 3 (11.05 g), and all three treatments in bioreactors were significantly higher than the explants cultured in a semi-solid medium, with 5.85 g. Similarly, for dry weight, all three treatments in bioreactors ($p < 0.001$) (1.18 g to 1.39 g) were significantly higher than the control cultured in a semi-solid medium (0.51 g); however, there were no significant differences among the dry weights of the three treatments.

Total numbers of shoots ($p < 0.004$) (15.4 to 17.5 shoots per explant) in bioreactors were significantly higher than those in the control group using a semi-solid medium, with 9.5 shoots per explant. The number of effective shoots ($p < 0.007$) (6.8 to 8.5 shoots per explant) in bioreactors was significantly higher than the control, with 3.9 shoots per explant.

The highest chlorophyll relative content was recorded in the control group in a semi-solid medium with an SPAD value of 22.0 ($p < 0.001$), followed by treatment 3 with an SPAD value of 14.8 ($p < 0.007$) and then treatment 1 and treatment 2 with SPAD values of 11.8 and 11.2, respectively (Figure 2).

3.2. Acclimatization Using IQ Forte as a Biostimulant

Survival rates varied greatly among the four groups. Treatments 1 (2.0 mL/L; 85%) and 2 (3.0 mL/L; 90%), where lower concentrations of the biostimulant were applied, had higher survival rates, while treatment 3 (4.0 mL/L; 70%) and the control group (75%) had lower survival rates (Figure 3).

For the plant growth data (including plant height, fresh weight, and dry weight), the differences between the three groups were not significant at the 0.05 level of significance according to ANOVA. At the same time, treatment 3 was significantly lower than the other two treatments, but it was not significantly different from the control group.

The highest plant height (16.1 cm) occurred in treatment 2, followed by treatment 1 (15.8 cm) and the control group (15.2 cm). Treatment 3 had a plant height of 12.9 cm, significantly lower than treatments 1 and 2 ($p < 0.02$). For fresh weight, treatments 1 and 2 (22.21 g to 20.48 g, respectively) were significantly higher than treatment 3 (11.17 g) ($p < 0.03$). There was no significant difference between the control group (16.24 g) and the other treatments. Similarly, for dry weight, treatment 3 (0.87 g) was significantly lower than treatments 1 (1.79 g) and 2 (1.61 g) ($p < 0.05$), with no significant difference between the control group (1.37 g) and the other groups (Figure 4).

For the relative chlorophyll content, results were slightly different. Treatment 3 was significantly lower than treatment 2 and the control, while treatment 1 was not significantly different from the other groups (Figure 4).

4. Discussion

The evaluation of the different in vitro culture systems demonstrated the usefulness of TI (temporary immersion) to increase the quantity and quality of shoots produced with respect to the conventional micropropagation system in a semi-solid medium [19]. Similarly, *Anthurium andreanum* Lind. and malanga (*Colocasia esculenta* L. Schott) multiplication can also be greatly improved in efficiency by TIS [19,20]. In studies with sugarcane and different types of TIS bioreactors, the efficiency of using temporary immersion systems for direct organogenesis has also been shown [21].

The two species mentioned above belong to the Araceae family, like *M. deliciosa*. In our study, *M. deliciosa* 'Thai Constellation' propagated in SETIS™ bioreactors achieved higher multiplication rates and increased fresh/dry weight as compared to those in a semi-solid culture medium. For the shoot multiplication rate, not only was the total number of new shoots significantly greater in temporary immersion bioreactors than in a semi-solid culture

medium, but the number of effective shoots produced by it was likewise much greater, which is of great significance for commercial production practice. Because each effective shoot that emerges can be used as a new single explant to enter the next round of multiplication, the effective shoot yield improvement during each round of multiplication will greatly affect the final production efficiency. The increase in fresh/dry weight in the bioreactor is also important. Shoots that accumulate more nutrients have a higher adaptation and survival rate during subsequent multiplication or acclimatization.

One of the main advantages of using TIS is that it promotes ventilation within the culture vessel, allowing the removal of volatile compounds such as ethylene [22], recirculating the carbon dioxide necessary for photosynthesis [23], and increasing stomatal functionality in the leaves compared to those obtained in semi-solid environments [24]. These factors could explain the increased efficiency of *M. deliciosa* multiplication in TIS.

Unlike the above parameters, however, for plant height, the data in the temporary immersion bioreactors were not fully ahead of the traditional semi-solid medium culture method. The average plant height in the control was not significantly different from either treatment 2 or treatment 3.

For the relative chlorophyll content, the experimental data collected contradicts many experiments. An increase in the content of photosynthetic pigments in TIS compared to a semi-solid system has been observed in malanga (*Colocasia esculenta*) [20]. In addition, an increase in chlorophyll synthesis has also been reported by using TIS in the micropropagation of *A. andreanum*, and the relative chlorophyll content was significantly higher for *Brassavola nodosa* (L.) Lindl. plantlets produced in TIS bioreactors as compared to those from a semi-solid medium [25]. In this experiment, however, in temporary immersion bioreactors, the relative chlorophyll content of the leaves of the explants was significantly lower than that of the control using the traditional method. The control group was significantly higher than all other treatments. Irradiance is an important factor in physiological processes that affects the synthesis of chlorophyll [26], leading to potentially two main reasons for our results:

The light system in the immersion bioreactors in this experiment was different from the lighting used in the control group. It has been shown that different light sources result in significant differences in relative chlorophyll content among banana varieties [10]. The effects of LED and fluorescent lamps were compared as sources of light on the growth and development of sweet basil

(*Ocimum basilicum* L.) and lemon balm (*Melissa officinalis* L.), indicating that the response of plants to the applied light is individual and depends on the species [27].

For micropropagated plants in the lab, photosynthesis is not really useful for their growth since we have already added a sufficient amount of sucrose to the medium as their nutrient source. Plant materials in TIS may instead accumulate relatively little chlorophyll due to higher growth rates.

Immersion and aeration frequency and duration in TIS bioreactors are factors that require adjustments for each species, and they play an important role in plant in vitro multiplication and plant development [28]. The optimization of such parameters is important to prevent physiological disorders such as hyperhydricity. Generally, the incidence of hyperhydricity increases due to the over-accumulation of water in plant tissues [29]. Two different immersion frequencies (1.5 h and 3 h) and two immersion durations (1 min and 2 min) were set to form three experimental groups in this experiment. For several parameters, such as dry weight, total new shoots produced, and number of effective shoots, the results of the three treatments were not significantly different. This is the first study on variegated *M. deliciosa* using TIS, and there were no previous reports to suggest optimum conditions for *Monstera* in general. Thus, this initial study explored some basic parameters for immersion using TIS bioreactors, warranting further studies to optimize the system, such as using shorter immersion duration and lower immersion frequency.

For plant height, treatment 1 with an immersion duration of 1 min was significantly higher than treatments 2 and 3 with an immersion duration of 2 min. At the time of experimental data collection, no significant hyperhydricity was observed. This suggests that a shorter immersion duration helps to increase the plant height of variegated *M. deliciosa*.

While the dry weights of the three treatments were not significantly different, the fresh weights of treatments 1 and 2 were significantly greater than those of treatment three. This implies that the water content of the plant material may be related to the frequency of immersion. A higher frequency of immersion would increase the water content of the culture. A similar trend has been described, where shorter immersion intervals gave rise to a higher number of shoots and a greater ratio of hyperhydric carnation plantlets cultured in the TIS [30].

The relative chlorophyll content of treatment 3 was significantly higher than that of treatments 1 and 2. This may indicate that in TIS, the relative chlorophyll content of the plant material is related to its water content.

The roles played by accumulated amino acids in plants vary, from acting as an osmolyte, regulating ion transport, modulating stomatal opening, and detoxifying heavy metals. Amino acids also affect the synthesis and activity of some enzymes, gene expression, and redox homeostasis [31]. By providing exogenous amino acids to maize seedlings, it was observed that in addition to proline, alanine, serine, and asparagine also delayed wilting under stress conditions [32]. In wheat, a foliar spray of methionine in a mixture increased drought tolerance and was associated with an increase in plant–water relations, physio-biochemical attributes, yield attributes, and nutrition quality when applied at 0.2 mg/mL [33]. Using two levels of methionine, i.e., 100 mg/L and 200 mg/L, on green parasol plants improved the absorption of N, P and K, which was associated with increases in dry weight of shoots, leaf area, and leaf chlorophyll contents [34]. In cowpea, a 0.4 mM concentration of methionine was effective in enhancing stress tolerance and improving growth, yield characteristics, contents of chlorophyll, carotenoids, shoot and seed nutrients, and other components [35].

Several studies mentioned above have demonstrated that a wide range of amino acid compositions have a beneficial effect on many species of plants, especially in terms of nutrient uptake and increasing their resistance to stress. The main components of the biostimulant used in this experiment are amino acids obtained from the hydrolysis of plant proteins, and we expected that they would play a significant role in the acclimatization of variegated *M. deliciosa*.

The results show that application of low concentrations of a biostimulant returned higher survival rates for variegated *M. deliciosa* seedlings, while high concentrations of a biostimulant had an opposite effect, leading to reduced survival rates.

In the experiment, the growth data of the plantlets (plant height, fresh weight, and dry weight) treated with low concentrations of biostimulants were not significantly different from the control. The reasons for this were analyzed as follows:

Insufficient sample number. This is mainly due to the high value of the material (variegated *M. deliciosa*) in this experiment, the high cost of expanding the sample numbers, and the lack of experience with its acclimatization process (death of plant material due to excessive sunlight and high temperatures). Subsequent experiments could increase the number of samples based on more mature acclimatization experience.

It is true that a proportion of plants are insensitive to biostimulants by means of increasing exogenous amino acids.

It is evident from the data that the use of low concentrations of a biostimulant did not significantly affect the relative chlorophyll content of leaves of variegated *M. deliciosa*. It is noteworthy that the SPAD values of treatment 3 were significantly lower than those of treatment 2 and the control, indicating that higher concentrations of amino acid solutions may, on the contrary, hinder the development of chloroplasts in the green parts of the leaves of variegated *M. deliciosa*.

5. Conclusions

This is the first study reporting the use of the SETIS™ bioreactor for the micropropagation of *M. deliciosa* 'Thai Constellation'. We determined that this variegated *M. deliciosa* under TIS bioreactors provided higher multiplication rates and increased fresh/dry weight compared to conventional culture systems using a semi-solid agar-based medium. Different immersion and aeration frequencies and duration in TIS bioreactors do affect the growth of the material in a number of ways, such as plant height, fresh weight, and chlorophyll content. Although the plant material in TIS has grown favorably under the conditions we set up, we still need to follow up with more experiments to find out if there are more suitable culture conditions for *M. deliciosa*. This is also the first study reporting the use of IQ Forte as a biostimulant for the acclimatization of *M. deliciosa* 'Thai Constellation'. We found that this biostimulant at low concentrations enhanced the acclimatization survival of variegated *M. deliciosa*. The gain in plantlets' growth was not significant, while higher concentrations of the biostimulant may have a negative effect on the growth of variegated *M. deliciosa*.

For future research, expanded experimentation is necessary. The first step could be to set up more different culture conditions, mainly different immersion and aeration intervals and durations, under conditions of more adequate plant material and more bioreactors. This setup of culture conditions could be based on this study to increase the differences in conditions to get more significant results. This study only proved the advantages of the TIS bioreactor in the multiplication of variegated *M. deliciosa*, and experiments on its rooting efficiency under the new system would be a good addition. This experiment did not demonstrate the effect of IQ Forte, the new biostimulant, well. The following directions can be taken for the subsequent improvement of the experiment: (1) Consider the effect of seasonal climate to provide more stable and safe acclimatization conditions for variegated *M. deliciosa* to avoid the occurrence of

stress. (2) Prepare sufficient experimental materials before the experiment. (3) Other cheaper varieties of *Monstera* can be used to test this novel biostimulant to reduce the cost of testing.

For commercial production, this study suggests the use of SETIS™ bioreactors instead of the system using a semi-solid agar-based medium at the stage of multiplication of *M. deliciosa* 'Thai Constellation'. The new system not only saves production space and labor but also expands the micropropagation of *M. deliciosa* at a higher efficiency.

Conceptualization, W.V.; methodology, W.V. and Y.J.; validation, W.V., Y.J. and D.B.; formal analysis, Y.J.; investigation, Y.J.; resources, W.V. and D.B.; data curation, Y.J.; writing—original draft preparation, Y.J.; writing—review and editing, W.V.; visualization, Y.J.; supervision, W.V. and D.B.; project administration, W.V. and D.B.; funding acquisition, W.V. All authors have read and agreed to the published version of the manuscript.

Not applicable.

Not applicable.

Data is contained within the article.

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Footnotes

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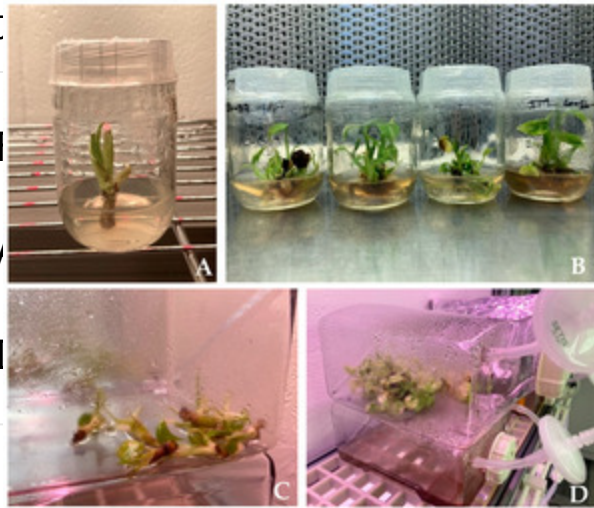


Figure 1. Micropropagation of *M. deliciosa* 'Thai Constellation' under different culture systems. (A,B) Semi-solid medium in baby food jar. (C,D) Liquid medium in SETIS™ temporary immersion bioreactors.

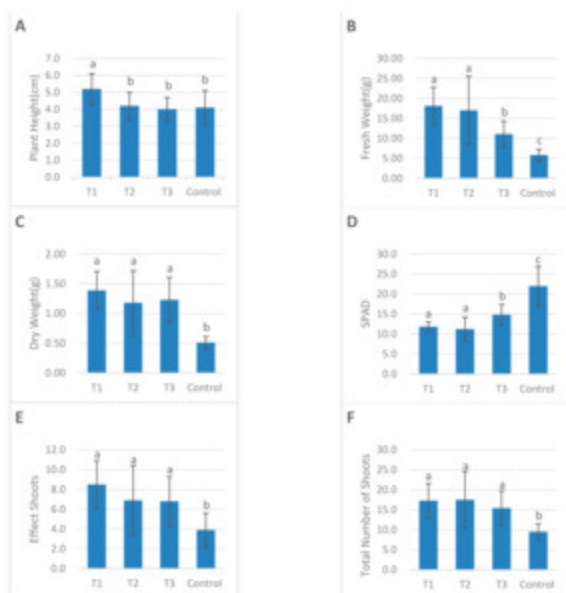


Figure 2. Effect of different in vitro culture systems on plants' development of *M. deliciosa* 'Thai Constellation' after 90 days culturing. (A) Plant height, (B) Fresh weight, (C) Dry weight, (D) SPAD, (E) Effective shoots, and (F) Total number of shoots. Treatment: Liquid medium in SETIS™ bioreactor; treatment 1: immersion frequency of 1.5 h, immersion duration of 1 min; treatment 2: immersion frequency of 1.5 h, immersion duration of 2 min; treatment 3: immersion frequency of 3 h, immersion duration of 2 min. Control: Semi-solid medium in baby food jar.

baby food jar. Bars indicate mean \pm SE. Different letters indicate significant differences by Tukey's test at $p \leq 0.05$.

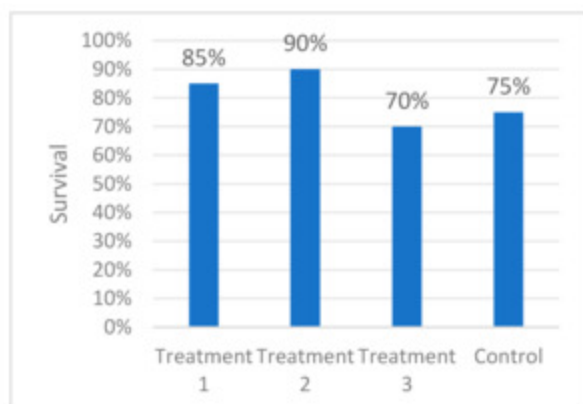


Figure 3. Survival of *M. deliciosa* 'Thai Constellation' after 100 days of growing in the greenhouse. Treatment 1: 2.0 mL/L biostimulant; treatment 2: 3.0 mL/L biostimulant; treatment 3: 4.0 mL/L biostimulant; control: water without biostimulant.

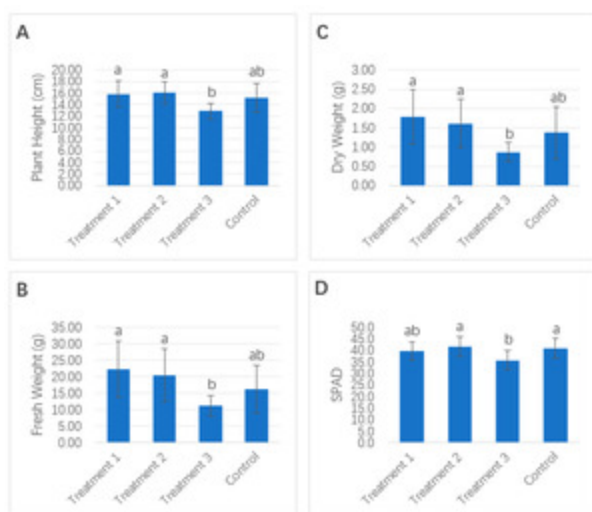


Figure 4. Effect of different concentrations of biostimulant on plants' development of *M. deliciosa* 'Thai Constellation' after 100 days of growing in the greenhouse. (A) Plant height, (B) Fresh weight, (C) Dry weight, (D) SPAD; treatment 1: 2.0 mL/L biostimulant; treatment 2: 3.0 mL/L biostimulant; treatment 3: 4.0 mL/L biostimulant; control: water

without biostimulant. Bars indicate mean \pm SE.

Different letters indicate significant differences by Tukey's test at $p \leq 0.05$.

References

1. Bailey, L.H.; Bailey, E.Z. *Hortus Third: A Concise Dictionary of Plants Cultivated in the United States and Canada*; MacMillan: New York, NY, USA, 1976; pp. 739-740.
2. Peppard, T.L. Volatile flavor constituents of *Monstera deliciosa*. *J. Agric. Food Chem.*; 1992; 40, pp. 257-262. [DOI: <https://dx.doi.org/10.1021/jf00014a018>]
3. Spínola, V.; Perestrelo, R.; Câmara, J.S.; Castilho, P.C. Establishment of *Monstera deliciosa* fruit volatile metabolomic profile at different ripening stages using solid-phase microextraction combined with gas chromatography–mass spectrometry. *Food Res. Int.*; 2015; 67, pp. 409-417. [DOI: <https://dx.doi.org/10.1016/j.foodres.2014.11.055>]
4. Dewei, H.A.N. Tissue Culture and Rapid Propagation of *Monstera deliciosa*. *J. Anhui Agric. Sci.*; 2010; 38, pp. 14270-14271.
5. Bhojwani, S.S.; Dantu, P.K.; Bhojwani, S.S.; Dantu, P.K. Micropropagation. *Plant Tissue Culture: An Introductory Text*; Springer: New Delhi, India, 2013; pp. 245-274.
6. Berthouly, M.; Etienne, H. Temporary immersion system: A new concept for use liquid medium in mass propagation. *Liquid Culture Systems for In Vitro Plant Propagation*; Springer: Dordrecht, The Netherlands, 2005; pp. 165-195.
7. Preil, W. General introduction: A personal reflection on the use of liquid media for in vitro culture. *Liquid Culture Systems for In Vitro Plant Propagation*; Springer: Dordrecht, The Netherlands, 2005; pp. 1-18.
8. Bello-Bello, J.J.; Cruz-Cruz, C.A.; Pérez-Guerra, J.C. A new temporary immersion system for commercial micropropagation of banana (*Musa* AAA cv. Grand Naine). *Vitr. Cell. Dev. Biol.-Plant*; 2019; 55, pp. 313-320. [DOI: <https://dx.doi.org/10.1007/s11627-019-09973-7>]
9. Ramírez-Mosqueda, M.A.; Bello-Bello, J.J. SETIS™ bioreactor increases in vitro multiplication and shoot length in vanilla (*Vanilla planifolia* Jacks. Ex Andrews). *Acta Phys. Plant*; 2021; 43, 52. [DOI: <https://dx.doi.org/10.1007/s11738-021-03227-z>]

10. Vendrame, W.A.; Feuille, C.; Beleski, D.; Bueno, P.M.C. In vitro growth responses of ornamental Bananas (*Musa* sp.) as affected by light sources. *Horticulturae*; 2022; 8, 92. [DOI: <https://dx.doi.org/10.3390/horticulturae8020092>]
11. Conover, C.A.; Poole, R.T. Acclimatization of indoor foliage plants. *Hort. Rev.*; 1984; 6, pp. 119-154.
12. Kozai, T. Acclimatization of micropropagated plants. *Biotech. Agric For.*; 1991; 17, pp. 127-141.
13. European Biostimulant Industry Council (EBIC). 2016; Available online: <http://www.biostimulants.eu/> (accessed on 23 October 2020).
14. Mrid, R.B.; Benmrid, B.; Hafsa, J.; Boukcim, H.; Sobeh, M.; Yasri, A. Secondary metabolites as biostimulant and bioprotectant agents: A review. *Sci. Total Environ.*; 2021; 777, 146204. [DOI: <https://dx.doi.org/10.1016/j.scitotenv.2021.146204>]
15. Schaafsma, G. Safety of protein hydrolysates, fractions thereof and bioactive peptides in human nutrition. *Eur. J. Clin. Nutr.*; 2009; 63, pp. 1161-1168. [DOI: <https://dx.doi.org/10.1038/ejcn.2009.56>]
16. Colla, G.; Nardi, S.; Cardarelli, M.; Ertani, A.; Lucini, L.; Canaguier, R.; Rouphael, Y. Protein hydrolysates as biostimulants in horticulture. *Sci. Hort.*; 2015; 196, pp. 28-38. [DOI: <https://dx.doi.org/10.1016/j.scienta.2015.08.037>]
17. Colla, G.; Hoagland, L.; Ruzzi, M.; Cardarelli, M.; Bonini, P.; Canaguier, R.; Rouphael, Y. Biostimulant action of protein hydrolysates: Unraveling their effects on plant physiology and microbiome. *Front. Plant Sci.*; 2017; 8, 2202. [DOI: <https://dx.doi.org/10.3389/fpls.2017.02202>] [PubMed: <https://www.ncbi.nlm.nih.gov/pubmed/29312427>]
18. Murashige, T.; Skoog, F. A revised medium for rapid growth and bioassays with tobacco tissue cultures. *Physiol. Plant*; 1962; 15, pp. 473-497. [DOI: <https://dx.doi.org/10.1111/j.1399-3054.1962.tb08052.x>]
19. Martínez-Estrada, E.; Islas-Luna, B.; Pérez-Sato, J.A.; Bello-Bello, J.J. Temporary immersion improves in vitro multiplication and acclimatization of *Anthurium andreanum* Lind. *Sci. Hort.*; 2019; 249, pp. 185-191. [DOI: <https://dx.doi.org/10.1016/j.scienta.2019.01.053>]

20. Arano-Avalos, S.; Gómez-Merino, F.C.; Mancilla-Álvarez, E.; Sánchez-Páez, R.; Bello-Bello, J.J. An efficient protocol for commercial micropropagation of malanga (*Colocasia esculenta* L. Schott) using temporary immersion. *Sci. Hort.*; 2020; 261, 108998. [DOI: <https://dx.doi.org/10.1016/j.scienta.2019.108998>]
21. Orozco-Ortiz, C.; Sanchez, L.; Araya-Matthey, J.; Vargas-Solorzano, I.; Araya-Valverde, E. BIT bioreactor increases in vitro multiplication of quality shoots in sugarcane (*Saccharum* spp. variety LAICA 04-809). *Plant Cell Tissue Org. Cult.*; 2022; 152, pp. 115-128. [DOI: <https://dx.doi.org/10.1007/s11240-022-02392-4>]
22. Roels, S.; Noceda, C.; Escalona, M.; Sandoval, J.; Canal, M.J.; Rodriguez, R.; Debergh, P. The effect of headspace renewal in a temporary immersion bioreactor on plantain (*Musa* AAB) shoot proliferation and quality. *Plant Cell Tissue Org. Cult.*; 2006; 84, pp. 155-163. [DOI: <https://dx.doi.org/10.1007/s11240-005-9013-y>]
23. Aragón, C.E.; Sánchez, C.; Gonzalez-Olmedo, J.; Escalona, M.; Carvalho, L.; Amâncio, S. Comparison of plantain plantlets propagated in temporary immersion bioreactors and gelled medium during in vitro growth and acclimatization. *Biol. Plant*; 2014; 58, pp. 29-38. [DOI: <https://dx.doi.org/10.1007/s10535-013-0381-6>]
24. Afreen, F. Temporary immersion bioreactor. *Plant Tissue Cult. Eng.*; 2008; 2008, pp. 187-201.
25. Vendrame, W.A.; Xu, J.; Beleski, D.G. Micropropagation of *Brassavola nodosa* (L.) Lindl. using SETIS™ bioreactor. *Plant Cell Tissue Org. Cult.*; 2023; 153, pp. 67-76. [DOI: <https://dx.doi.org/10.1007/s11240-022-02441-y>]
26. Mancilla-Álvarez, E.; Pérez-Sato, J.A.; Núñez-Pastrana, R.; Spinoso-Castillo, J.L.; Bello, J.J.B. Comparison of Different Semi-Automated Bioreactors for In vitro Propagation of Taro (*Colocasia esculenta* L. Schott). *Plants*; 2021; 10, 1010.
27. Frąszczak, B.; Golcz, A.; Zawirska-Wojtasiak, R.; Janowska, B. Growth rate of sweet basil and lemon balm plants grown under fluorescent lamps and LED modules. *Acta Sci. Pol. Hortorum Cultus*; 2014; 13, pp. 3-13.
28. Georgiev, V.; Schumann, A.; Pavlov, A.; Bley, T. Temporary immersion systems in plant biotechnology. *Eng. Life Sci.*; 2014; 14, pp. 607-621. [DOI: <https://dx.doi.org/10.1002/elsc.201300166>]

29. Saher, S.; Piqueras, A.; Hellin, E.; Olmos, E. Hyperhydricity in micropropagated carnation shoots: The role of oxidative stress. *Physiol. Plant.*; 2004; 120, pp. 152-161. [DOI: <https://dx.doi.org/10.1111/j.0031-9317.2004.0219.x>] [PubMed: <https://www.ncbi.nlm.nih.gov/pubmed/15032887>]

30. Ahmadian, M.; Babaei, A.; Shokri, S.; Hessami, S. Micropropagation of carnation (*Dianthus caryophyllus* L.) in liquid medium by temporary immersion bioreactor in comparison with solid culture. *J. Gen. Eng. Biotechnol.*; 2017; 15, pp. 309-315. [DOI: <https://dx.doi.org/10.1016/j.jgeb.2017.07.005>] [PubMed: <https://www.ncbi.nlm.nih.gov/pubmed/30647668>]

31. Rai, V.K. Role of amino acids in plant responses to stresses. *Biol. Plant.*; 2002; 45, pp. 481-487. [DOI: <https://dx.doi.org/10.1023/A:1022308229759>]

32. Thakur, P.S.; Rai, V.K. Exogenously supplied amino acids and water deficits in *Zea mays* cultivars. *Biol. Plant.*; 1985; 27, pp. 458-461. [DOI: <https://dx.doi.org/10.1007/BF02894717>]

33. Hammad, S.A.; Ali, O.A. Physiological and biochemical studies on drought tolerance of wheat plants by application of amino acids and yeast extract. *Ann. Agric. Sci.*; 2014; 59, pp. 133-145. [DOI: <https://dx.doi.org/10.1016/j.aosas.2014.06.018>]

34. Shekari, G.; Javanmardi, J. Effects of foliar application pure amino acid and amino acid containing fertilizer on broccoli (*Brassica oleracea* L. var. *italica*) transplants. *Adv. Crop Sci. Technol.*; 2017; 5, pp. 280-287. [DOI: <https://dx.doi.org/10.4172/2329-8863.1000280>]

35. Merwad, A.R.M.; Desoky, E.S.M.; Rady, M.M. Response of water deficit-stressed *Vigna unguiculata* performances to silicon, proline or methionine foliar application. *Sci. Hort.*; 2018; 228, pp. 132-144. [DOI: <https://dx.doi.org/10.1016/j.scienta.2017.10.008>]


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Abstract

Translate

Monstera deliciosa Liebm. 'Thai Constellation' is a variegated variety of *M. deliciosa* belonging to the family Araceae, which has become a new favorite in the foliage plant market. However, limited studies exist on its propagation, and growers have difficulties in achieving large-scale production. This study aimed at developing an efficient protocol for the micropropagation of *M. deliciosa* using SETIS™ temporary immersion bioreactors. Furthermore, we aimed at evaluating the role of a novel biostimulant (IQ Forte) in the acclimatization of *M. deliciosa*. Significant differences were observed among the different treatments, showing higher multiplication rates under TIS conditions as compared to the semi-solid control. Adjusting immersion parameters also showed benefits in improving multiplication rates. The novel biostimulant (IQ Forte) did not provide significant gains in growth of *M. deliciosa* 'Thai Constellation' during acclimatization.

Details

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