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Accelerated Dragon Fruit Production through Integrated Tissue Culture and Micro-Grafting Techniques

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ABSTRACT

Dragon fruit (*Hylocereus spp*.) is a high value crop crucial for meeting rising global demand; however, its commercial expansion is hampered by slow propagation, high pathogen susceptibility, and abiotic stresses such as drought and salinity. To address these challenges, we developed an integrated propagation protocol that combines optimized *in vitro* tissue culture with advanced micro grafting techniques to produce disease free, genetically uniform plantlets with enhanced vigor and resilience. Tissue culture experiments were performed using a meticulously formulated Murashige and Skoog medium (pH 5.7–5.8) supplemented with 7 g/L agar, 30 g/L sucrose, and cytokinins; benzylaminopurine (BAP), metatopolin (MT), and kinetin (KIN) at concentrations ranging from 0.5 to 1.5 mg/L. Notably, treatment with 1.5 mg/L MT yielded 5.8 shoots per explant, promoted 4.5 cm shoot elongation, achieved a 93% survival rate, and reduced emergence time to 8.9 days. Concurrent micro grafting trials paired Palora cv. DF1 (yellow) scions with Vitname white cv. as DF2 (white) and Lisa cv. as DF3 (red) rootstocks high yielding, achieving grafting success rates above 88%. Histological analyses revealed superior vascular reconnection and tissue integration, which translated into enhanced photosynthetic performance (12.5 µmol CO m^2 s^1) and robust biomass accumulation. This scalable, sustainable protocol not only accelerates propagation and early fruiting but also markedly improves plant vigor and adaptability, offering a robust framework for revolutionizing commercial dragon fruit cultivation in semi arid and tropical regions. Future research will focus on long-term field validation, genetic stability, and economic feasibility.

Keywords: *Hylocereusspp*- Plant growth regulators (PGRs)-Metatopolin- Shoot proliferation-Biomass accumulation.

INTRODUCTION

Dragon fruit (*Hylocereus spp*.), also known as pitaya, is a captivating cactus species cultivated for its vibrantly colored fruit with a refreshingly sweet and subtly floral flavor. Native to Central and South America, dragon fruit has gained global popularity due to its unique appearance, nutritional benefits, and medicinal properties. The increasing market demand for this exotic fruit necessitates efficient propagation methods to ensure sustainable production and meet commercial requirements (Xu and Wang, 2024 and Heikel et al., 2025).Dragon fruit has become economically significant, particularly in tropical and subtropical regions, with major producers including Vietnam, China, Mexico, Ecuador, and Thailand (Mordor Intelligence, 2024). Its high market value and adaptability to diverse climatic conditions make it an attractive crop for semi-arid regions, including Egypt, where financial evaluations indicate that dragon fruit farming is non-capital intensive, with relatively low investment costs compared to annual production expenses (Elshishtawy, 2024).

Traditional propagation of dragon fruit through stem cuttings is widely practiced but suffers from several limitations, including slow multiplication rates, which restricting large scale production, disease transmission risks, such as stem canker, bacterial soft rot, and fungal infections (Yadav et al., 2025). Inability to propagate genetically superior cultivars or stress tolerant rootstocks, soil related issues, including salinity, poor drainage, and climate-induced drought stress (Anushi and Ghosh, 2024 and

Lakshmeshwara *et al*., 2024). To overcome these limitations, tissue culture emerges as a revolutionary technique, offering rapid multiplication, disease-free plantlets, and genetic uniformity (Khezri *et al*., 2024 and Mishra *et al*., 2024).

Tissue culture emerges as a revolutionary technique to overcome the limitations associated with conventional propagation methods. Also known as micro-propagation, it involves the *invitro* culture of plant cells, tissues, or organs on a sterilized nutrient medium within a controlled laboratory environment. This method offers several advantages for dragon fruit propagation, including rapid multiplication, production of disease-free plantlets, and the ability to propagate elite cultivars with desirable traits. Furthermore, tissue culture is an effective tool in breeding programs; facilitating genetic improvement and conservation efforts (George et al., 2007and George et al., 2008).At the cellular level, plant growth regulators (PGRs) play a critical role in tissue culture by inducing cell differentiation and plant regeneration. Auxins and cytokinins regulate organogenesis, stimulating root and shoot formation, respectively (Loyola-Vargas andOcho-Alejo, 2018).

Building upon the advantages of tissue culture, micro-grafting presents another innovative approach for enhancing dragon fruit production. Micro-grafting involves grafting a desired scion cultivar onto a well-established rootstock with superior characteristics such as disease resistance, drought tolerance and improved vigor (Wang et al., 2022). The success of this technique depends on the formation of vascular tissue connections, which facilitate the efficient transport of water, nutrients, and signaling molecules between the scion and rootstock. Proper vascular alignment ensures physiological integration, supporting uniform growth, enhanced productivity, and improved resilience (Singh et al., 2024). One of the most significant advantages of micro-grafting in dragon fruit cultivation is its ability to reduce the juvenile phase, leading to earlier fruit production. Certain rootstocks, such as Megalinces, have been reported to shorten the juvenile phase of the scion cultivar, accelerating the transition to reproductive maturity (Borchetia *et al*., 2022 and Manchanda *et al*., 2022). This is particularly beneficial for commercial growers seeking higher yields in shorter timeframes.

While the individual benefits of tissue culture (Xu and Wang, 2024) and micro-grafting (Arya *et al*., 2024) in dragon fruit propagation are well documented, comprehensive research on their integrated application remains limited. Traditional propagation via stem cuttings is plagued by slow multiplication rates and a heightened risk of disease transmission including stem canker, bacterial soft rot, and fungal infections which constrain large scale production (Wakchaure et al., 2021 andMedemba et al., 2012). In contrast, tissue culture using Murashige and Skoog medium supplemented with optimal concentrations of plant growth regulators has been demonstrated to produce rapid, disease-free plantlets with high genetic stability (Xu and Wang., 2024). Similarly, micro-grafting enables the effective combination of desirable scion traits with robust, drought-tolerant rootstocks, thereby improving overall plant vigor and adaptability (Arya et al., 2024 and Janick and Paull, 2008).

By integrating these methodologies, researchers and growers can harness the synergistic advantages of both approaches to maximize propagation efficiency and enhance crop resilience under stress conditions. This research aims to develop an efficient tissue culture protocol for the rapid multiplication of ‘white, red, and yellow dragon fruit variety and evaluate the compatibility and graft success rate when micro-grafting the ‘yellow dragon fruit scion onto the ‘white and red dragon fruit’ rootstocks; The outcomes are expected to establish a scalable, sustainable propagation strategy for the commercial production of disease-resistant and high-performing dragon fruit cultivars.

MATERIALS AND METHODS

1. Experimental sites:

This study was conducted on 2023- 2024 cross two international sites, the Horticulture Research Institute (HRI), Agricultural Research Center (ARC), Giza, Egypt. And Tissue Culture and Laboratory Plant of the Cryopreservation Environmental Horticulture Department, Institute of Food and Agricultural Sciences (IFAS), University of Florida (UF), Gainesville, USA.

2. Plant material:

Actively growing, healthy shoots were collected from certified, disease-free mother plants representing three dragon fruit (*Hylocereus spp*.) cultivars: Palora ‘DF1’ (*H. megalanthus*, yellow skin), Vietnam White ‘DF2’ (*H. undatus*, white flesh), and Lisa ‘DF3’ (*H. costaricensis*, red flesh). Explants measuring 1.5 –2.5 cm (average 2.0 cm) with multiple nodes were selected to ensure uniformity and optimal physiological status for *invitro* propagation (Arunkumar *et al*., 2022 and Mustafa *et al*., 2021).

3. Surface sterilization:

Collected explants were first washed thoroughly under running tap water to remove surface debris. Surface disinfection was then carried out under aseptic conditions in a laminar flow hood using a sequential protocol; immersion in 1% sodium hypochlorite solution for 20 minutes with gentle agitation, brief rinse in 70% ethanol for 3–5 seconds, then three successive rinses with sterile distilled water to remove residual sterilants (Majid *et al*., 2014).

4. Culture medium preparation:

Murashige and Skoog (MS) basal medium (Murashige and Skoog., 1962) was used for all tissue culture experiments. The medium was adjusted to pH 5.7–5.8 prior to autoclaving. Medium contained 30 g/L of sucrose and 7 g/L of agar. Media were prepared under sterile conditions and supplemented with the following plant growth regulators (PGRs):

5- Benzylaminopurine (BAP): 0.5, 1.0, and 1.5 mg/L.

Meta-Topolin [6-(3-hydroxybenzylamino) purine] (MT): 0.5, 1.0, and 1.5 mg/L.

Kinetin (KIN): 0.5, 1.0, and 1.5 mg/L

A control group consisting of MS medium without PGRs was also included. Each treatment was clearly labeled and replicated for statistical validation.

6. *InVitro* propagation and micro-grafting:

6.1. Micro propagation protocol:

Sterilized explants were cultured on MS media supplemented with the above PGR concentrations. Cultures were maintained at 25 ± 2°C under a 16-hour photoperiod. Parameters such as shoot proliferation rate, number of shoots per explant, and survival percentage were recorded after four weeks of incubation.

6.2. Micro-grafting procedure:

Micro-grafting was carried out using the pasting method under aseptic conditions. The scion Palora cultivar (‘DF1’ yellow dragon fruit) was grafted onto two *invitro* propagated rootstocks Vitname white cultivar (‘DF2’ white-fleshed and Lisa cultivar ‘DF3’ red-fleshed dragon fruit) derived from tissue culture. Rootstocks were 4–6 weeks old from subculture, with well-developed, turgid cladode segments approximately 4–5 cm in length and 0.5–0.8 cm in diameter, ensuring active meristamatic growth. Scions were excised from healthy donor plants of similar physiological age, trimmed to match the cut surface of the rootstock, and gently pressed into place without mechanical fixation, relying on the natural mucilage for adhesion. Grafted plantlets were immediately transferred to a controlled-environment greenhouse and maintained under high relative humidity (>85%), 25 ±2 °C air temperature, and diffused light (PPFD 80–100 µmol m^2s^1) for four weeks to promote graft union formation. Graft success and scion–rootstock compatibility were assessed at 4 weeks post-grafting by recording survival percentage and measuring early vegetative growth parameters.

7. Morphological and biomass evaluations:

7.1. *InVitro* evaluations: After 4 weeks in culture, the following parameters were recorded:

Morphological and growth parameters:

Shoot proliferation: Number of shoots per explant, counted manually.

Shoot growth: Length of the shoots (cm) measured with a calibrated ruler.

Explants survival: Expressed as the percentage of explants that remained viable over the culture period.

Rooting characteristics:

Rooting percentage: Proportion of explants that developed roots.

Number of roots/ explant: Counted for each explant.

Root length: Average length of roots (cm) measured with a digital caliper.

Biomass accumulation and emergence:

Fresh and dry weight: Determined by weighing plantlets immediately after harvest and after oven-drying at 70°C for 48 hours, respectively.

Time to shoot emergence: Recorded as the number of days until the first shoot became visible.

7.2. Micro-grafting evaluations

For the micro-grafted plantlets, evaluations were performed during the post-grafting acclimatization phase after 2 weeks as follows:

Grafting success rate (%): Calculated as the percentage of grafted explants that formed a successful union.

Time to graft union formation (days): Recorded as the number of days until a visible graft union was established.

Visual assessment of graft union: The quality of the union was evaluated on a scale of 1–5, based on callus formation and the establishment of vascular continuity.

Biomass and physiological analyses: Fresh and dry weights of the grafted plants were recorded to provide an index of overall plant vigor.

Biochemical analyses: Leaf tissue, collected at the onset of flowering, was used to determine:

Chlorophyll content (mg/g FW): Measured spectrophotometrically at 645 and 663 nm (Amoo *et al*., 2011).

Total carbohydrates (mg/g FW): Determined using the phenol-sulfuric acid method (Dubios et al., 1956).

Total phenolic compounds (mg/g FW): Quantified using the Folin–Ciocalteu reagent method (Nemes et al., 2018).

7.3. Physiological, hormonal, and graft union formation evaluations:

To further elaborate on the impact of graft union formation and overall plant health, the following additional measurements were incorporated:

Photosynthetic parameters: Fully expanded healthy cladode segments were used to measure net photosynthetic rate (µmol CO m^2 s^1) and stomatal conductance (mmol HO m^2 s^1) using a portable photosynthesis system (LI-COR 6400). Measurements were taken on flat mid-segments, normalized to cuvette area, and recorded after stabilization (<5% drift in 60 s). Instruments were calibrated daily, and five biological replicates per treatment were sampled from standardized canopy positions. These methods followed standard operating protocols (Long and Bernacchi, 2003 and Flexas et al., 2008).

Leaf (cladode) area (cm^2): The projected surface area of the cladode (the flattened, photosynthetic stem) was measured by the portable leaf area meter (LI-3000C).

Root-to-shoot ratio: was calculated by dividing the dry root weight by the dry shoot weight (Cornelissen et al., 2003).

Hormone profiling: Leaf tissues collected at the onset of flowering were analyzed for hormone content. Auxin (IAA) and cytokinin (measured as Zeatin) levels were quantified using high-performance liquid chromatography (HPLC) based on the protocol described by (Grossmann, 2010).

8. Statistical analysis:

All experiments were arranged in a completely randomized design (CRD) with three replications per treatment to minimize variability among experimental units (Gomez and Gomez., 1984). Data were statistically analyzed using IBM SPSS Statistics version 22.0. A one-way analysis of variance (ANOVA) was performed, followed by the Least Significant Difference (LSD) test to detect significant differences among treatment means at P<0.05. In addition, Partial Least Squares Path Modeling (PLS-PM) was conducted using Smart PLS 4.0 to quantify the causal relationships between the different propagation strategies and physiological adaptation to stress. This integrated approach allowed us to not only compare treatments statistically but also to explore the underlying interactions between key experimental factors.

Fig.(1). Photographic Documentation of the Initial Stages of the Propagation Protocol.

RESULTS

1. Effect of cytokinins on shooting and growth performance:

The application of different cytokinins (BAP, MT, and KIN) at varying concentrations significantly influenced *invitro* shoot proliferation, shoot elongation, and survival rates in white, red, and yellow dragon.

1.1. White Dragon fruit:

For white dragon fruit, the results revealed that shoot proliferation, shoot length, and survival percentage were significantly influenced by the type and concentration of plant growth regulators (PGRs) applied **(Table, 1A).** The application of Meta-topolin (MT) at 1.5 mg/L resulted in the highest proliferation rate (5.8 ± 0.3 shoots/explant), shoot length (4.5 ± 0.4 cm), and survival percentage (93 ± 1%), with all values showing statistically significant superiority ($p < 0.05$) over most other treatments. BAP at 1.0 mg/L also performed well, yielding 5.3 ± 0.2 shoots/ explant and 4.2 ± 0.3 cm shoot length with a survival rate of 91 ± 2%, which was not significantly different from BAP at 1.5 mg/L or MT at 1.0 mg/L. Conversely, the use of KIN at 0.5 mg/L resulted in the lowest shoot proliferation (4.0 ± 0.3), shortest shoots (3.4 ± 0.3cm), and lowest survival (89 ± 3%), indicating limited efficacy for *invitro* regeneration of white dragon fruit at this concentration. Generally, MT at 1.5 mg/L proved to be the most effective PGR for promoting vigorous and healthy plant development in white dragon fruit under *invitro* conditions.

Table (1A). Effect of different plant growth regulators and their concentrations on *invitro* shoot proliferation; shoot growth, and survival of white dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Proliferation (shoots/explant)	Shoot Growth (cm)	Survival (%)
BAP	0.5	4.5 ± 0.3 bc	3.8 ± 0.4 bc	91 ± 2 ab
BAP	1	5.3 ± 0.2 ab	4.2 ± 0.3 ab	91 ± 2 ab
BAP	1.5	5.0 ± 0.2 ab	4.2 ± 0.3 ab	92 ± 2 ab
MT	0.5	4.2 ± 0.3 c	3.6 ± 0.4 c	90 ± 2 ab
MT	1	4.8 ± 0.2 bc	4.0 ± 0.3 bc	92 ± 2 ab
MT	1.5	5.8 ± 0.3 a	4.5 ± 0.4 a	93 ± 1 a
KIN	0.5	4.0 ± 0.3 c	3.4 ± 0.3 c	89 ± 3 b
KIN	1	4.6 ± 0.2 bc	3.9 ± 0.3 bc	91 ± 2 ab
KIN	1.5	4.8 ± 0.2 bc	4.0 ± 0.4 bc	90 ± 2 ab

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

1.2. Red Dragon fruit:

Application of cytokinins significantly influenced *invitro* shoot proliferation, elongation, and survival in red dragon fruit as it shown in **(Table, 1B).** Among the tested treatments, Meta-topolin (MT) at 1.5 mg/L resulted in the highest proliferation rate (5.4 shoots/ explant), maximum shoot length (4.3 cm), and superior survival percentage (92%), all statistically significant compared to other treatments. BAP at 1.5 mg/L and MT at 1.0 mg/L also demonstrated enhanced shoot development, although lower than MT at 1.5 mg/L. In contrast, KIN at 0.5 mg/L showed the lowest performance across all parameters. These findings highlight MT, particularly at 1.5 mg/L, as the most effective cytokinin for improving micro-propagation efficiency in white dragon fruit.

Table (1B). Effect of different plant growth regulators and their concentrations on *invitro* shoot proliferation; shoot growth, and survival of red dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Proliferation (shoots/explant)	Shoot Growth (cm)	Survival (%)
BAP	0.5	4.0 ± 0.3 c	3.4 ± 0.4 c	88 ± 2 b
BAP	1	4.7 ± 0.3 b	3.9 ± 0.4 b	90 ± 2 ab
BAP	1.5	4.9 ± 0.3 ab	4.1 ± 0.4 ab	89 ± 2 ab
MT	0.5	4.3 ± 0.3 bc	3.7 ± 0.4 bc	90 ± 2 ab
MT	1	5.1 ± 0.2 ab	4.1 ± 0.4 ab	92 ± 2 a
MT	1.5	5.4 ± 0.3 a	4.3 ± 0.4 a	92 ± 2 a
KIN	0.5	3.7 ± 0.3 c	3.2 ± 0.3 c	87 ± 2 b
KIN	1	4.4 ± 0.2 bc	3.6 ± 0.3 bc	89 ± 2 ab
KIN	1.5	4.6 ± 0.3 bc	3.7 ± 0.3 bc	88 ± 2 b

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

1.3. Yellow Dragon fruit:

(Table, 1C) presents the effects of three PGRs (BAP, MT, and KIN) at concentrations of 0.5, 1, and 1.5mg/L on *invitro* shoot proliferation, shoot growth, and survival of yellow dragon fruit after 4 weeks. For shoot proliferation, both BAP and MT at 1.5 mg/L (5.0 ± 0.3 and 5.1 ± 0.2, respectively) significantly outperformed lower concentrations, while KIN at 0.5 mg/L (3.7 ± 0.3) yielded the lowest proliferation. Intermediate values were observed for BAP and MT at 1.0 mg/L (both around 4.9) and for KIN at 1.0 and 1.5 mg/L (4.3-4.5), suggesting a clear dose-dependent response. Shoot elongation followed a similar trend; BAP at 1.5 mg/L produced the longest shoots (4.2 ± 0.4 cm), and MT at 1.0 mg/L also reached optimal shoot length (4.2 ± 0.3 cm), whereas both KIN and MT at 0.5 mg/L resulted in the shortest shoots (3.1–3.3 cm). Survival rates were consistently high with BAP and MT (around 90–91%), while KIN at 0.5 mg/L showed a slightly lower survival rate (87%). Data indicated that BAP and MT, particularly at concentrations between 1.0 and 1.5 mg/L, significantly enhance shoot proliferation and elongation as well as maintain high survival in white dragon fruit compared to KIN.

Table (1C). Effect of different plant growth regulators and their concentrations on *invitro* shoot proliferation; shoot growth, and survival of yellow dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Proliferation (shoots/explant)	Shoot Growth (cm)	Survival (%)
BAP	0.5	4.1 ± 0.3 d	3.5 ± 0.4 c	89 ± 2 b
BAP	1	4.9 ± 0.3 b	4.0 ± 0.4 b	91 ± 1 a
BAP	1.5	5.0 ± 0.3 a	4.2 ± 0.4 a	90 ± 2 a
MT	0.5	4.0 ± 0.3 d	3.3 ± 0.3 d	88 ± 2 b
MT	1	4.9 ± 0.2 b	4.2 ± 0.3 a	91 ± 2 a
MT	1.5	5.1± 0.2 a	4.0 ± 0. 3 b	91 ± 2 a
KIN	0.5	3.7 ± 0.3 e	3.1 ± 0.3 d	87 ± 3 c
KIN	1	4.3 ± 0.2 c	3.5 ± 0.3 c	89 ± 2 b
KIN	1.5	4.5 ± 0.2 c	3.6 ± 0.3 c	88 ± 2 b

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

2. Effect of cytokinins on rooting performance:

Significant differences (*P*< 0.05) were observed in rooting percentage, number of roots per explant, and root length across all cytokinin treatments and dragon fruit varieties **(Tables 2A–2C).** The response to the different plant growth regulators varied not only by PGR type and concentration but also by genotype.

2.1. White Dragon Fruit:

Data in **(Table, 2A)** illustrated that, Meta-topolin (MT) at 0.5 mg/L resulted in the highest rooting percentage (81.2%), the greatest number of roots per explant (3.8), and the longest root length (5.1 cm). This was followed by BAP and KIN at 0.5 mg/L, which also supported high rooting rates of 76.5% and 73.0%, respectively. Increasing the concentration of any cytokinin beyond 1.0 mg/L led to a significant reduction in all rooting traits, with BAP at 1.5 mg/L producing the poorest rooting percentage (45.20 %) and shortest root length (2.3 cm). These results suggest that lower cytokinin concentrations are more favorable for rooting in white dragon fruit, with MT showing superior performance across all traits.

Table (2A). Effect of cytokinin type and concentration on rooting performance of white dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Rooting (%)	Number of Roots/Explant	Root Length (cm)
BAP	0.5	76.5 ± 2.3 ab	3.3 ± 0.2 ab	4.6 ± 0.2 ab
BAP	1	61.5 ± 2.1 d	2.5 ± 0.2 cd	3.6 ± 0.2 d
BAP	1.5	45.2 ± 2.4 f	1.8 ± 0.2 e	2.3 ± 0.2 f
MT	0.5	81.2 ± 2.0 a	3.8 ± 0.2 a	5.1 ± 0.3 a
MT	1	68.9 ± 2.4 c	2.8 ± 0.2 c	4.0 ± 0.3 c
MT	1.5	54.1 ± 1.9 e	2.1 ± 0.2 d	2.9 ± 0.3 e
KIN	0.5	73.0 ± 1.8 b	3.1 ± 0.3 b	4.2 ± 0.3 bc
KIN	1	60.2 ± 2.0 d	2.3 ± 0.3 cd	3.3 ± 0.2 d
KIN	1.5	48.7 ± 2.2 f	19 ± 0.3 de	2.5 ± 0.2 f

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

2.2. Red Dragon Fruit:

The red cultivar followed a similar trend as it shown in **(Table, 2B),** with MT at 0.5 mg/L out performing all other treatments, achieving the highest rooting percentage (77.6%), number of roots (3.6), and root length (4.9 cm). KIN and BAP at 0.5 mg/L also supported relatively high rooting rates (69.4% and 71.9%, respectively), although statistically lower than MT. As in the white cultivar, increasing cytokinin concentrations led to diminished performance, particularly with BAP and KIN at 1.5 mg/L. Notably, MT consistently showed a positive effect on root development**,** indicating its effectiveness across different Hylocereus genotypes.

Table (2B). Effect of cytokinin type and concentration on rooting performance of red dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Rooting (%)	Number of Roots/Explant	Root Length (cm)
BAP	0.5	71.9 ± 2.0 ab	3.2 ± 0.3 ab	4.5 ± 0.3 a
BAP	1	57.3 ± 2.1 d	2.4 ± 0.2 d	3.4 ± 0.2 d
BAP	1.5	42.7 ± 2.4 f	1.6 ± 0.2 f	2.0 ± 0.2 f
MT	0.5	77.6 ± 2.1 a	3.6 ± 0.2 a	4.9 ± 0.2 a
MT	1	64.8 ± 2.0 c	2.7 ± 0.2 c	3.8 ± 0.2 cd
MT	1.5	50.6 ± 1.9 e	2.0 ± 0.2 e	2.7 ± 0.2 e
KIN	0.5	69.4 ± 2.2 bc	3.0 ± 0.2 bc	4.1 ± 0.2 bc
KIN	1	55.9 ± 2.3 d	2.2 ± 0.3 de	3.0 ± 0.2 e
KIN	1.5	44.1 ± 2.1 f	1.7 ± 0.2 f	2.2 ± 0.3 f

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

3.2.3. Yellow Dragon Fruit:

The yellow cultivar also showed the best rooting response under MT at 0.5 mg/L, with a rooting rate of (75.8%), root number of (3.4), and root length of 4.6 cm as it described in **(Table, 2C).** Treatments with KIN and BAP at the same concentration followed closely. The negative effect of increasing cytokinin concentration was also observed in this genotype, with rooting rates declining significantly at 1.5 mg/L, particularly under BAP (40.9%). While KIN was slightly more consistent than BAP at higher concentrations, MT remained the most effective cytokinin in promoting rooting and overall root system development.

Table (2C). Effect of cytokinin type and concentration on rooting performance of yellow dragonfruit after 4 weeks.

PGR	Concentration (mg/L)	Rooting (%)	Number of Roots/Explant	Root Length (cm)
BAP	0.5	70.1 ± 2.1 ab	3.0 ± 0.2 ab	4.2 ± 0.2 ab
BAP	1	56.9 ± 2.3 d	2.3 ± 0.2 cd	3.1 ± 0.2 cd
BAP	1.5	40.9 ± 2.4 f	1.7 ± 0.2 e	2.0 ± 0.2 f
MT	0.5	75.8 ± 2.0 a	3.4 ± 0.2 a	4.6 ± 0.2 a
MT	1	63.3 ± 2.2 c	2.6 ± 0.2 c	3.5 ± 0.3 c
MT	1.5	49.3 ± 2.1 e	2.0 ± 0.2 d	2.5 ± 0.2 e
KIN	0.5	67.8 ± 2.0 b	2.9 ± 0.s3 b	3.8 ± 0.2 b
KIN	1	55.5 ± 2.1 d	2.2 ± 0.2 cd	2.8 ± 0.3 d
KIN	1.5	43.6 ± 2.3 f	1.8 ± 0.2 e	2.1 ± 0.2 f

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

2.4. Comparative Overview:

Across all three cultivars, MT at 0.5 mg/L consistently led to superior rooting performance**,** indicating its broad applicability for micro-propagation of dragon fruit. In contrast, BAP at 1.5 mg/L repeatedly resulted in the lowest values for all rooting parameters. This highlights the dose-dependent and genotype-dependent effects of cytokinins**,** with lower concentrations

3. Biomass accumulation in *InVitro*-propagated Dragon fruit explants:

Application of different cytokinin types and concentrations significantly influenced fresh and dry biomass accumulation as well as the timing of shoot emergence in white dragon fruit plantlets.

3.1. White Dragon Fruit:

Data in **(Table, 3A)** indicates that fresh weight was highest under 0.5 mg/L KIN (3.12 g) and 0.5 mg/L MT (3.10 g), with statistically significant differences compared to lower-performing treatments (BAP at 1.0 and 1.5 mg/L). Moreover, dry weight was significantly greater in explants treated with 0.5 mg/L BAP (0.58 g) and 1.5 mg/L KIN (0.57 g), while MT at 1.0 and 1.5 mg/L resulted in lower biomass accumulation. While, **Table (3A)** illustrated that, time to shoot emergence varied from 8.9 to 10.1 days. The earliest emergence was observed under BAP and MT at 1.0 and 1.5 mg/L, whereas BAP at 1.5 mg/L showed a significantly delayed response.

Table (3A). Effect of cytokinin treatments on biomass accumulation and shoot emergence in white dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Fresh Weight (g)	Dry Weight (g)	Time to Shoot Emergence (days)
BAP	0.5	2.90 ± 0.15 ab	0.58 ± 0.05 a	9.1 ± 0.2 ab
BAP	1	2.77 ± 0.15 b	0.53 ± 0.05 ab	8.9 ± 0.2 b
BAP	1.5	2.93 ± 0.15 ab	0.53 ± 0.05 ab	10.1 ± 0.2 a
MT	0.5	3.10 ± 0.15 ab	0.56 ± 0.05 a	9.4 ± 0.2 ab
MT	1	2.75 ± 0.15 b	0.45 ± 0.05 b	9.5 ± 0.2 ab
MT	1.5	2.75 ± 0.15 b	0.46 ± 0.05 b	8.9 ± 0.2 b
KIN	0.5	3.12 ± 0.15 a	0.52 ± 0.05 ab	9.3 ± 0.2 ab
KIN	1	2.95 ± 0.15 ab	0.50 ± 0.05 ab	9.5 ± 0.2 ab
KIN	1.5	2.71 ± 0.15 b	0.57 ± 0.05 a	9.0 ± 0.2 b

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

3.2.Red Dragon Fruit:

The application of cytokinins significantly influenced fresh and dry biomass accumulation and the time to shoot emergence in red dragon fruit plantlets as it shown in **(Table, 3B).** Among the tested treatments, Meta-topolin (MT) at 1.0 mg/L exhibited the highest fresh weight (3.46 g)and dry weight (0.64 g)**,** with a shorter time to shoot emergence (9.1 days)**,** indicating superior shoot initiation and metabolic activity. Conversely, KIN at 1.5 mg/L resulted in the lowest fresh and dry weights (2.69 g and 0.53 g, respectively)and a delayed emergence (10.3 days), suggesting diminished physiological performance at higher Kinetin levels. While all treatments with MT showed relatively strong performance, the MT 1.0 mg/L combination proved statistically superior in most parameters ($P \leq 0.05$), making it the most efficient for improving shoot vigor and propagation speed.

Table (3B). Effect of cytokinin treatments on biomass accumulation and shoot emergence in red dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Fresh Weight (g)	Dry Weight (g)	Time to Shoot Emergence (days)
BAP	0.5	3.09 ± 0.15 ab	0.61 ± 0.02 ab	9.6 ± 0.3 ab
BAP	1	2.85 ± 0.12 bc	0.50 ± 0.02 c	9.8 ± 0.2 ab
BAP	1.5	2.93 ± 0.14 bc	0.53 ± 0.03 bc	10.5 ± 0.3 a
MT	0.5	2.85 ± 0.13 bc	0.61 ± 0.03 ab	10.2 ± 0.2 a
MT	1	3.46 ± 0.16 a	0.64 ± 0.02 a	9.1 ± 0.2 b
MT	1.5	3.00 ± 0.15 ab	0.61 ± 0.03 ab	10.2 ± 0.3 a
KIN	0.5	2.74 ± 0.13 c	0.59 ± 0.02 bc	9.8 ± 0.3 ab
KIN	1	3.21 ± 0.15 ab	0.58 ± 0.02 bc	9.7 ± 0.2 ab
KIN	1.5	2.69 ± 0.12 c	0.53 ± 0.02 c	10.3 ± 0.3 a

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

3.3. Yellow Dragon Fruit:

The cytokinin treatments exhibited notable and distinct effects on yellow dragon fruit's biomass accumulation and shoot emergence as it shown in **(Table, 3C).** Specifically, BAP at 0.5 and 1.0 mg/L maximized fresh weight (2.65 g), while increasing BAP to 1.5 mg/L significantly reduced fresh biomass (2.37 g). In contrast, MT showed a moderate, dose-dependent increase in fresh weight (ranging from 2.45 g to 2.65 g), and KIN consistently resulted in lower fresh biomass (2.33–2.47 g). For dry weight, both BAP and KIN at 1.5 mg/L achieved the highest accumulation (0.55–0.56 g), with lower concentrations yielding reduced values, and MT peaking modestly at 1.0 mg/L (0.54 g). Regarding shoot emergence, BAP at 1.5 mg/L significantly accelerated emergence (9.4 days), whereas BAP at 1.0 mg/L delayed it (11.9 days) and KIN at 1.0 mg/L promoted earlier emergence (9.8 days).

Table (3C). Effect of cytokinin treatments on biomass accumulation and shoot emergence in yellow dragon fruit after 4 weeks.

PGR	Concentration (mg/L)	Fresh Weight (g)	Dry Weight (g)	Time to Shoot Emergence (days)
BAP	0.5	2.65 a	0.45 c	11 bc
BAP	1	2.64 a	0.53 ab	11.9 e
BAP	1.5	2.37 b	0.55 a	9.4 a
MT	0.5	2.45 b	0.5 b	11.5 d
MT	1	2.55 ab	0.54 a	11.1 bc
MT	1.5	2.65 a	0.51 b	10.8 bc
KIN	0.5	2.43 b	0.47 c	11.1 bc
KIN	1	2.47 b	0.51 b	9.8 ab
KIN	1.5	2.33 b	0.56 a	10.9 bc

Values are means ± SE (n=5). Different letters in the same column indicate significant differences at P ≤ 0.05 (LSD test).

Fig.2 (A-H), Fig. 3(A-C) and Fig. 4(A-E) presented the comparative morphological responses of dragon fruit explant to different cytokinin treatments. **Fig. 2 (A-H)** illustrates the effects of increasing Kinetin concentrations (0.5, 1.0, and 1.5 mg/L), revealing a gradual improvement in shoot induction and elongation as concentration increases, although the overall uniformity and vigor remain moderate. In **Fig. 3 (A-C),** the impact of Benzylaminopurine (BAP) is depicted; here, the explants exhibit enhanced shoot proliferation and improved architecture at higher concentrations (1.0 and 1.5 mg/L) compared to the lowest dose, indicating BAP's efficacy in promoting shoot development. In contrast, **Fig. 4 (A-E)** demonstrates the superior performance of Meta-topolin (MT) at 1.5 mg/L, where shoot clusters exhibit remarkable density, uniformity, and elongation.

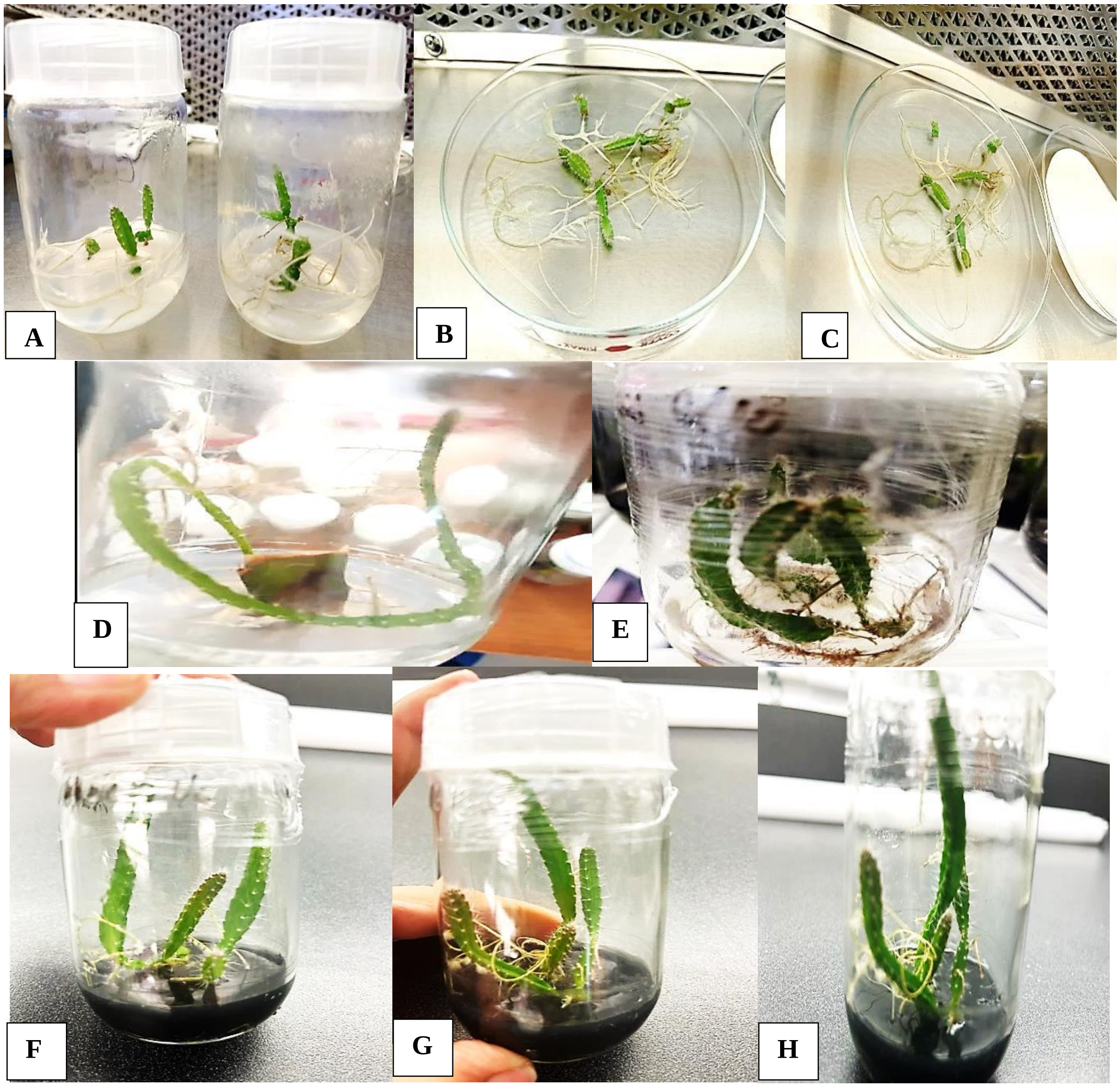


Fig. 2 (A-H). Comparative morphological effects of Kinetin treatments (0.5, 1.0, and 1.5 mg/l) on dragon fruit explants.

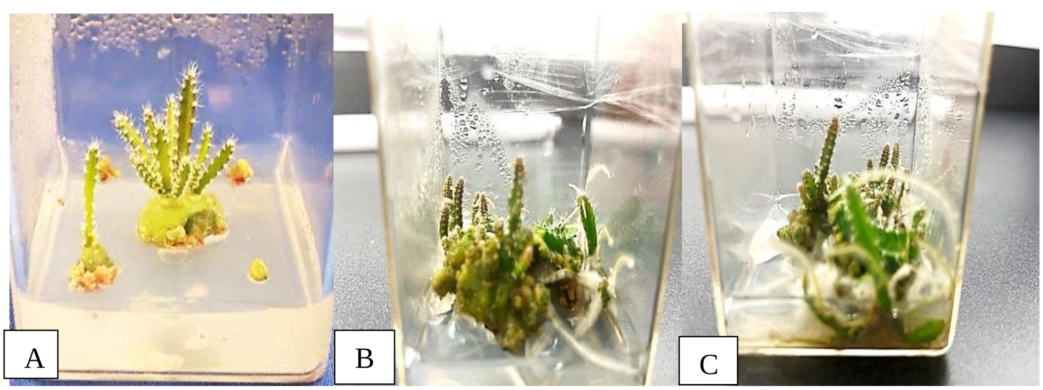


Fig. (3). (A-C). Comparative morphological effects of Benzylaminopurine (BAP) treatments (0.5, 1.0, and 1.5 mg/l) on dragon fruit explants.

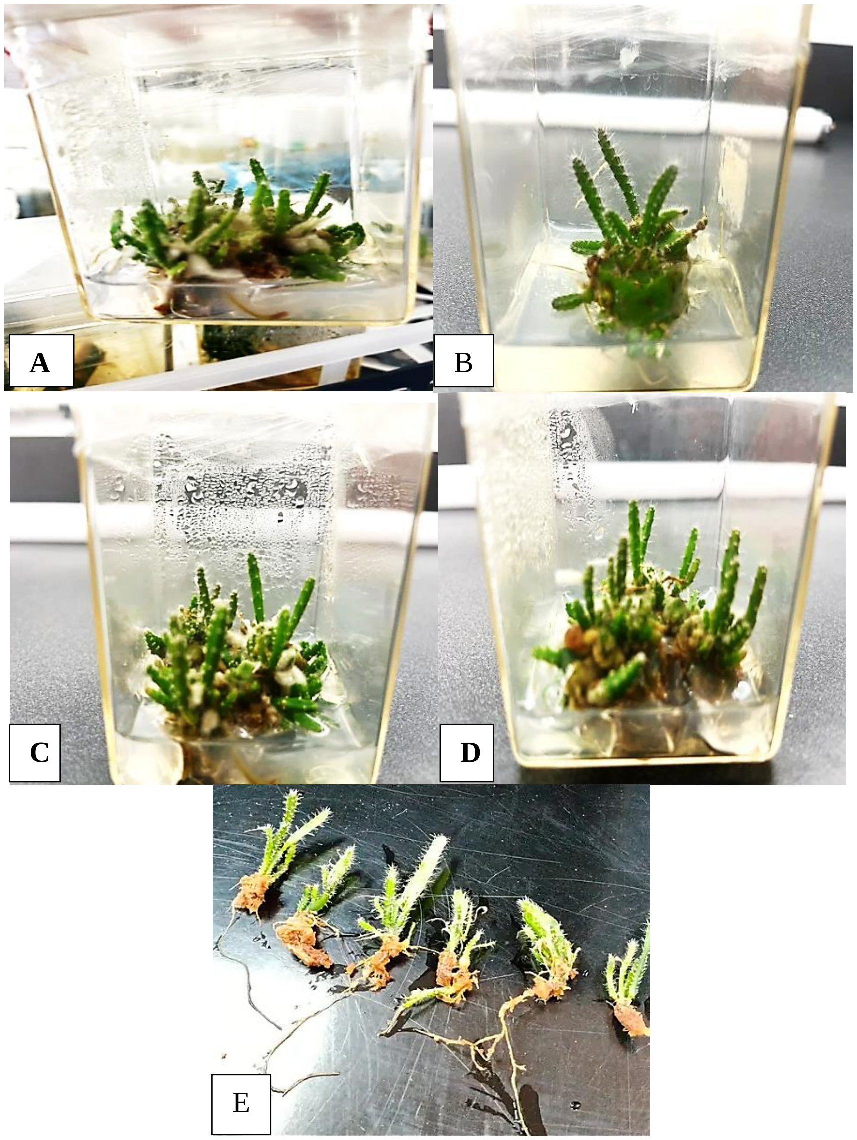


Fig. 4 (A-E). Comparative morphological effects of Meta-topolin (MT) treatments (0.5, 1.0, and 1.5 mg/l) on dragon fruit explants.

4.Micro-grafting efficiency and graft union quality in dragon fruit plantlets:

4.1.Graft union formation:

Grafting success, time to union, visual assessment scores, and microscopic evaluation of vascular reconnection and tissue integration were assessed as it is presented in **Fig. (5 A to L).**The micro-grafting experiment revealed distinct differences in graft union formation and subsequent plant performance between the two scion–rootstock combinations. The DF1/DF2 (White) combination which demonstrated in **Fig.(5A)** exhibited a higher grafting success rate (92.5%) compared to DF1/DF3 (Red) (88.0%), indicating a superior overall compatibility when using the DF2 rootstock. Although the DF1/DF3 combination formed the graft union faster (10.8 days versus 12.3 days) for DF1/DF2, the visual quality of the graft union as assessed on a 1–5 scale was significantly higher for DF1/DF2 (4.8) than for DF1/DF3 (4.5). This trend was further confirmed by histological evaluations, where DF1/DF2 plantlets showed a higher percentage of vascular reconnection (85%) and a superior tissue integration score (4.8) compared to DF1/DF3 (78% and 4.5, respectively), suggesting that the quality of the graft union and the establishment of effective vascular continuity are enhanced in the DF1/DF2 combination as described it **(Fig. 5 A, B, C).**

4.2.Biomass accumulation and biochemical indicators:

Fresh and dry weight measurements, chlorophyll content, total carbohydrates, and phenolic compounds as indices of plant vigor and metabolic status. Data shown in **(Fig. 5D)** of biomass accumulation and physiological performance indicated that, DF1/DF2 treatment also showed advantages. The fresh weight of DF1/DF2 plantlets averaged 15.2 ± 0.8 g, which was significantly greater than the (14.0 g) observed in DF1/DF3, although the dry weights (4.8 g) compared with (4.5 g) were not significantly different. Additionally, in (Fig. 5E), chlorophyll content which is indicative of photosynthetic capacity, was higher in DF1/DF2 (2.10 mg/g FW) relative to DF1/DF3 (1.95 mg/g FW). Higher levels of total carbohydrates were also noted in DF1/DF2, suggesting better energy storage and overall plant vigor, while total phenolic content remained statistically similar between treatments.

4.3.Photosynthetic performance and biomass allocation

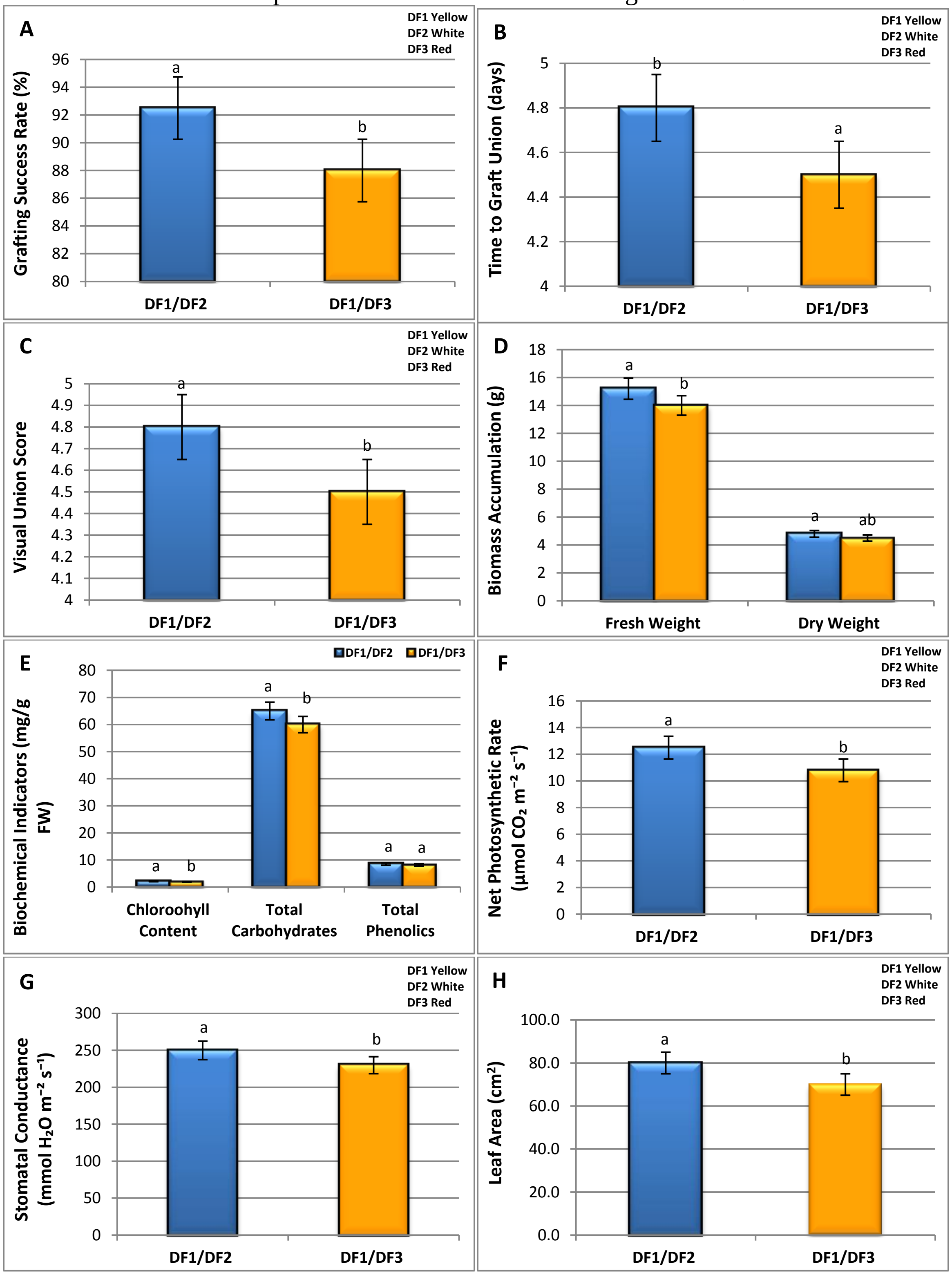
Physiological parameters including the photosynthetic rate, stomatal conductance, leaf area, and root-to-shoot ratio, which collectively reflect gas exchange efficiency and biomass partitioning. Further physiological assessments underscored these differences. DF1/DF2 plantlets recorded a higher photosynthetic rate (12.5 µmol CO_2 m^{-2} s^{-1}) in (Fig. 5F*)*, and stomatal conductance (250 mmol H_2O m^{-2} s^{-1}) in (Fig. 5G) compared to DF1/DF3 (10.8 µmol CO_2 m^{-2} s^{-1} and 230 mmol H_2O m^{-2} s^{-1}, respectively), as well as a larger leaf area (80.0 cm^2 vs. 70.0 cm^2) in (Fig. 5H). The root-to-shoot ratio in **(Fig. 5I),** which provides insight into biomass allocation, was lower for DF1/DF2, indicating proportionally greater shoot development a desirable trait in grafted plantlets destined for further cultivation.

4.4.Hormonal profiling and role in graft union formation

Hormonal analyses in **(Fig. 5J)** focusing on auxin (IAA) and cytokinin (Zeatin) levels to illustrate their influence on cell elongation, division, and overall graft compatibility. Hormone profiling further complemented these physiological insights. Data in Fig. 5D presented that DF1/DF2 plantlets exhibited higher auxin (IAA) concentrations (3.20 ng/g FW) compared to DF1/DF3 (2.80 ng/g FW), which likely contributed to enhanced cell elongation and vascular differentiation essential for graft union formation. Conversely, cytokinin levels (measured as Zeatin) were marginally higher in DF1/DF3 (2.00 ng/g FW) than in DF1/DF2 (1.80 ng/g FW), a factor that may promote cell division; however, in this context, the overall hormonal balance in DF1/DF2 appears to favor better anatomical integration as supported by the histological data.

4.5.Integrated visual assessment of graft union quality in micro-grafted dragon fruit:

(Fig. 5K) illustrate the subjective visual parameters of the graft union, including callus formation and vascular continuity as seen by human eye, which in turn combine to produce an overall visual quality score. The DF1/DF2 combination consistently shows higher scores compared to DF1/DF3, suggesting better graft union quality. Moreover, Findings in **(Fig. 5L)** complemented the visual assessment with objective histological measurements. Here, DF1/DF2 exhibits a higher percentage of vascular reconnection and a superior tissue integration score, reinforcing the conclusion that this combination produces a more robust graft union.



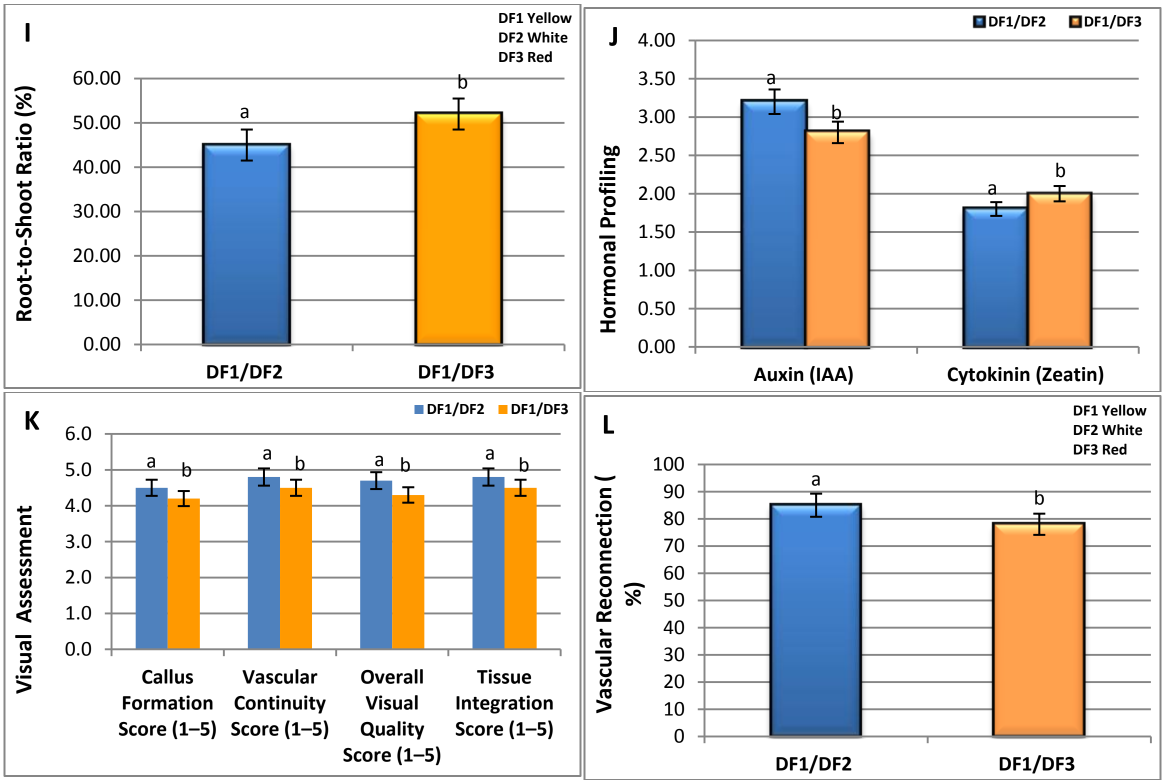
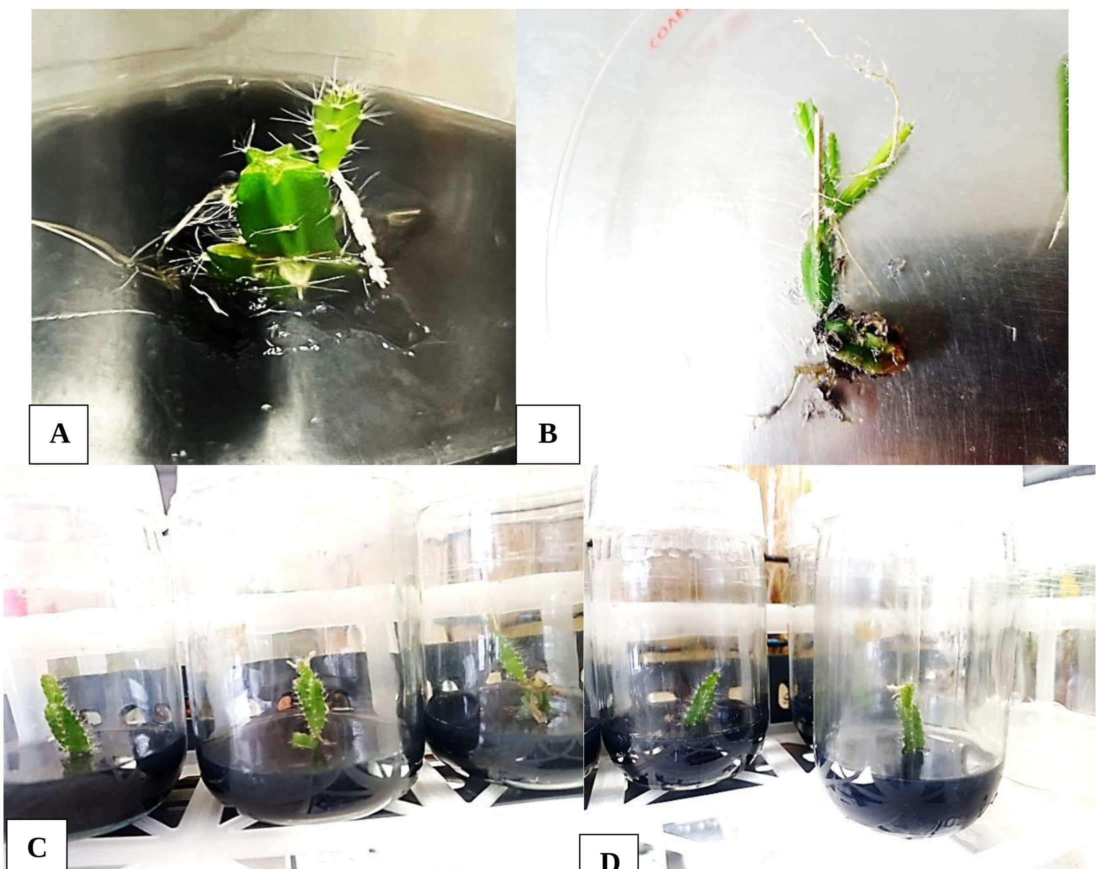


Fig. 5 (A- L). Micro-grafting efficiency and graft union quality assessments in dragon fruit plantlets.



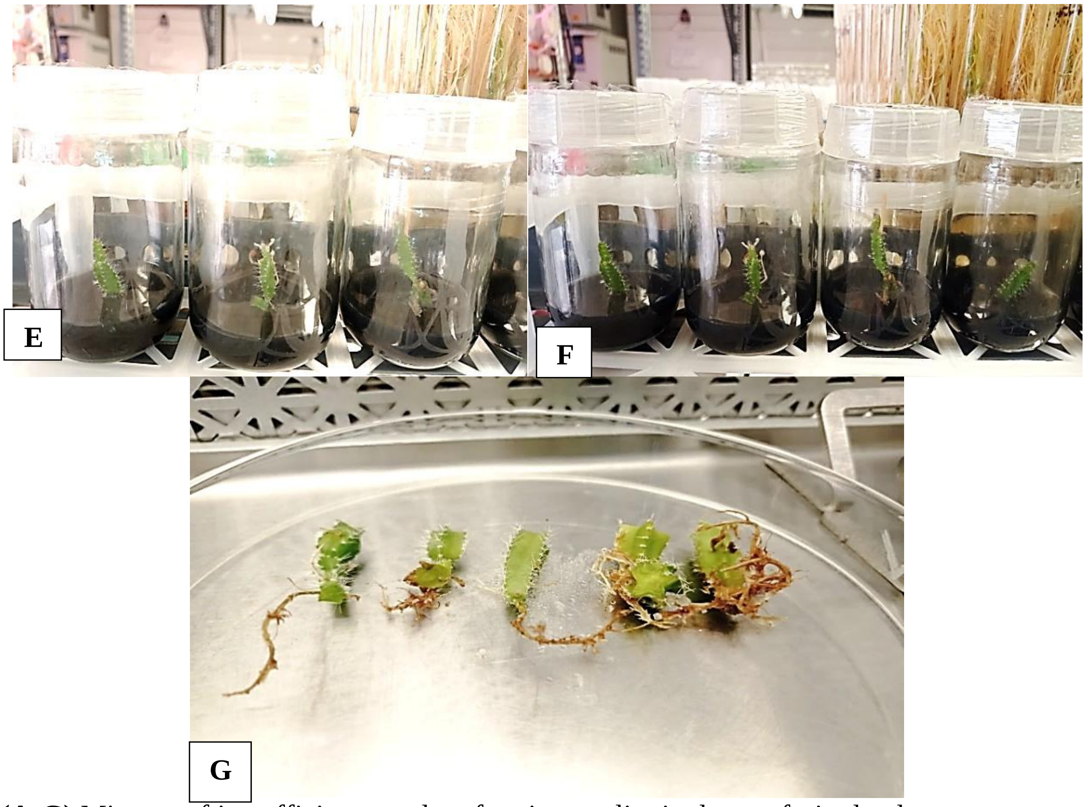


Fig. 6 (A-G). Micro-grafting efficiency and graft union quality in dragon fruit plantlets.

DISCUSSION

The present study demonstrates that an integrated propagation system combining optimized tissue culture with advanced micro-grafting techniques can effectively address the limitations inherent in conventional dragon fruit (*Hylocereus* spp.) propagation. Traditional methods, often reliant on stem cuttings, suffer from slow multiplication rates, high risks of pathogen transmission, and pronounced sensitivity to abiotic stresses such as drought and salinity (Wakchaure et al., 2021 and Yadav et al., 2025). Our approach, by contrast, yields disease-free, genetically uniform plantlets with enhanced vigor and resilience, thereby offering a scalable solution for commercial production in both semi-arid and tropical regions (Elshishtawy, 2024 and UNEP, 2023).

1. Optimized tissue culture conditions and cytokinin physiology:

Our *invitro* experiments highlighted the critical importance of optimizing both the nutrient medium and the type/concentration of plant growth regulators (PGRs). The precisely formulated Murashige and Skoog (MS) medium (pH 5.7–5.8, 7 g/ L agar, and 30 g /L sucrose) provided a stable nutritional environment for explants. Among the cytokinins evaluated benzylaminopurine (BAP), kinetin (KIN), and meta-topolin (MT) treatment with 1.5 mg L^{1} MT emerged as the most effective, producing 5.8 shoots per explant, average shoot elongation of 4.5 cm, a survival rate of 93%, and a reduced emergence time of 8.9 days.

The physiological basis for MT's superiority lies in its aromatic structure and metabolism. MT is less prone to causing hyperhydricity and oxidative stress than BAP, and it sustains meristematic activity over a longer period. It also maintains a more favorable cytokinin:auxin ratio, which supports both shoot elongation and auxin-mediated root initiation. BAP, while a potent stimulator of cell division, can at higher concentrations disrupt hormonal balance, leading to shorter shoots and reduced rooting. KIN generally has lower receptor affinity and uptake efficiency in succulent tissues, which may explain its comparatively weaker morphogenic response. These differences in uptake, receptor binding, and downstream hormonal balance explain the variation in proliferation, elongation, and survival rates observed among the three cytokinins (Amoo et al., 2011, Bouzroud et al., 2022, Krishna et al., 2021, Koç, 2021, Mishra et al., 2024and Xu and Wang, 2024).

2. Micro-grafting synergy and scion–rootstock compatibility:

Our micro-grafting experiments underscored the pivotal role of scion–rootstock interactions in determining graft success and subsequent plant performance. Pairing DF1 (yellow) scions with DF2 (white) and DF3 (red) rootstocks yielded success rates above 88%, but DF1/DF2 was markedly superior, with a 92.5% success rate, higher visual union scores, and improved vascular reconnection (85% vs. 78%) confirmed histologically (Ruzin, 1999).

Physiologically, this advantage can be explained by closer anatomical congruence between DF1 and DF2 including similar cladode thickness, vascular bundle arrangement, and cambial activity rates which facilitates precise alignment of xylem and phloem at the graft interface. This structural match accelerates callus proliferation, cambial bridge formation, and functional vascular continuity, enabling more efficient transport of water, minerals, and photo-assimilates (Moraless et al., 2023 and Wang et al., 2022). Hormonal signaling also plays a role: DF1/DF2 grafts showed elevated indole-3-acetic acid (IAA) levels, indicating more effective auxin transport from scion to rootstock. Auxin is critical for stimulating cambial differentiation and vascular strand formation, so this enhanced hormonal integration likely underpins the higher photosynthetic rate (12.5 µmol CO m^2s^1), greater chlorophyll content, and stronger biomass accumulation observed in DF1/DF2 compared to DF1/DF3. In DF1/DF3, subtle mismatches in vascular architecture or hormone transport efficiency may have slowed integration, reducing physiological performance under both optimal and stress conditions.

3. Physiological adaptation:

The enhanced performance of DF1/DF2 grafts under simulated salinity and drought stress reflects the combined benefits of anatomical precision and hormonal compatibility. Efficient vascular reconnection ensures stable water and nutrient supply, while balanced hormone signaling particularly sustained auxin flow supports stomatal regulation, carbon assimilation, and continuous xylem development under stress (Grossmann, 2010 and Loyola-Vargas and Ocho-Alejo, 2018). This integration allows the grafted plants to maintain high photosynthetic rates and biomass accumulation even in adverse environments; aligning with previous findings those strong graft unions improve stress resilience through coordinated hydraulic and hormonal function.

4. Sustainability, scalability and future perspectives:

By shortening the propagation cycle and reducing land and water inputs per plantlet, this integrated protocol aligns with circular bio-economy principles and supports sustainable intensification in water-limited regions (UNEP, 2023). The combination of MT optimized tissue culture and anatomically compatible grafting offers a reproducible, high-throughput system for producing vigorous, stress-resilient *Hylocereus* plants. Future research should focus on multi season field validation, long term genetic stability across propagation cycles, and economic feasibility analyses to confirm commercial scalability.

In summary**,** the physiological advantages conferred by MT during micro-propagation, coupled with the anatomical and hormonal compatibility of optimal scion–rootstock pairings, underpin the enhanced vigor, stress tolerance, and productivity observed. This integrated approach represents a significant advance in dragon fruit propagation science and offers a robust framework for both research and industry adoption (George et al., 2007 and Janich and Paull, 2008).

CONCLUSION

This study shows that integrating optimized tissue culture with advanced micro grafting markedly enhances dragon fruit propagation. A precisely formulated MS medium, adjusted to pH 5.7–5.8 with 7 g/L agar and 30 g/L sucrose, combined with optimal cytokinins (BAP, MT, KIN) enabled rapid multiplication of disease free, genetically stable plantlets. Notably, MT treatment outperformed the others by enhancing shoot proliferation, elongation, and survival rate. Concurrent micro grafting, which paired high yield scions achieved grafting success rates over 88% and ensured superior vascular reconnection and tissue integration, as verified by histological analysis. These improvements translated into enhanced photosynthetic performance and robust biomass accumulation. Overall, this integrated approach overcomes the limitations of traditional propagation methods and offers a scalable, sustainable solution for commercial dragon fruit production. Future work should focus on validating long term field performance, genetic stability, and economic feasibility to further refine this promising technique.

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الملخص العربي

اسراع إنتاج فاكهة التنين من خلال زراعة الأنسجة المتكاملة وتقنيات التطعيم الدقيق

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1 قسم بحوث الفاكهة الاستوائية، معهد بحوث البساتين - مركز البحوث الزراعية، الجيزة، مصر.

2 قسم البستنة البيئية - معهد علوم الأغذية والزراعة - جامعة فلوريدا، جينزفيل، فلوريدا، الولايات المتحدة الأمريكية.

تُعدّ ثمرة التنين (*Hylocereus spp.*) من المحاصيل عالية القيمة، وتكتسب أهمية متزايدة في ظل الطلب العالمي المتنامي. غير أن توسع زراعتها التجاري يواجه تحديات رئيسية أبرزها بطء معدلات التكاثر، وارتفاع حساسيتها لمسببات الأمراض وتأثرها الشديد بالظروف البيئية غير الحيوية مثل الجفاف والملوحة .وللتغلب على هذه التحديات، طوّرنا بروتوكولًا متكاملًا للإكثار يجمع بين زراعة الأنسجة المُحسّنة في المعمل وتقنيات التطعيم الدقيق المتقدمة بهدف إنتاج شتلات خالية من الأمراض، ومتجانسة وراثيًا وذات قوة ومرونة محسّنة.أُجريت تجارب زراعة الأنسجة باستخدام وسط موراشيج وسكوج (MS)مُحضّر بعناية (درجة حموضة 5.7–5.8)، مدعّم بـ 7 جم/لتر أجار، و30 جم/لتر سكروز، ومجموعة من السيتوكينينات شملت البنزيل أمينوبورين(BAP) ، والميتاتوبولين(MT) ، والكينيتين (KIN) بتركيزات تراوحت بين 0.5 و1.5 ملجم/لتر. وقد أظهرت المعاملة بتركيز 1.5 ملجم/لتر من المادة النشطة أفضل النتائج، حيث أنتجت 5.8 براعم لكل عينة، وعزّزت استطالة البراعم بمقدار 4.5 سم، وحققت معدل بقاء بلغ 93%، مع تقليص فترة الإنبات إلى 8.9 يوم .وفي تجـارب التطعيم الدقيق المتزامنة، جرى تطعيم صنف (*Palora DF1*الأصفر) على أصناف عالية المحصول مثل (*Vietnam White*الأبيض) و (*Lisa*الأحمر)، محققًا نسب نجاح تجاوزت 88%. وأظهرت التحاليل النسيجية إعادة اتصال وعائي وتكاملًا نسيجيًا متميزًا، انعكس في تحسين كفاءة التمثيل الضوئي (12.5 ميكرومول/CO_2 م$^{-2}$·ثانية) وزيادة تراكم الكتلة الحيوية .يمثل هذا البروتوكول القابل للتطوير والمستدام خطوة نوعية لا تُسرّع فقط من عملية الإكثار والإثمار المبكر، بل تعزز أيضًا قوة النبات وقدرته على التكيف، مما يضع أساسًا متينًا لإحداث نقلة نوعية في زراعة فاكهة التنين التجارية بالمناطق شبه القاحلة والاستوائية. وستركز الأبحاث المستقبلية على التحقق الميداني طويل الأمد، وضمان الاستقرار الوراثي، وتقييم الجدوى الاقتصادية.