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Effect of bio-regulators on grafting success of Macadamia nut (*Macadamia integrifolia* Maiden and Betché)

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ABSTRACT

The present investigation entitled “Effect of bio-regulators on grafting success of Macadamia nut (*Macadamia integrifolia* Maiden and Betché)” was carried out at Varashree Nursery, Shivamogga district, Karnataka, during 2025-2026 to study the effect of bio-regulators on graft success and survivability of Macadamia nut. The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications comprising fourteen treatments, viz., T_0 - Control (No dip), T_1 - Water dip, T_2 - NAA @ 10 ppm, T_3 - NAA @ 20 ppm, T_4 - NAA @ 30 ppm, T_5 - BAP @ 10 ppm, T_6 - BAP @ 20 ppm, T_7 - BAP @ 30 ppm, T_8 - GA_3 @ 5 ppm, T_9 - GA_3 @ 10 ppm, T_{10} - GA_3 @ 15 ppm, T_{11} - Biovita @ 2500 ppm, T_{12} - Biovita @ 5000 ppm and T_{13} - Biovita @ 7500 ppm. Among the treatments, the scion treated with (T_4) NAA @ 30 ppm, recorded the highest sprouting percentage (65.84%), graft success (64.04%), graft survivability (63.78%) and maximum scion girth (9.10 mm), whereas T_9 (GA_3 @ 10 ppm) recorded the minimum days taken for sprouting (22.86 days) and maximum scion length (37.68 cm) at 150 days after grafting. The findings indicated that scion treated with NAA @ 30 ppm was effective for graft establishment and survivability, while GA_3 @ 10 ppm promoted early sprouting and scion elongation compared to the other treatments.

Keywords: Macadamia nut, Grafting, Bio-regulators, NAA, GA_3 , Graft success

Introduction

Macadamia (*Macadamia integrifolia* Maiden and Betché) belongs to the family Proteaceae and is an important nut crop of the world. It is indigenous to the rainforests of north-eastern Australia and has subsequently been introduced into several tropical and subtropical regions. Major commercial development of macadamia has taken place in Hawaii, particularly during the last six decades (Sharma *et al.*, 2015) [1]. At present, it is commercially cultivated in the countries such as Australia, Brazil, Indonesia, Kenya, New Zealand and South Africa. Macadamia nuts are highly valued for their distinctive buttery flavour, crisp texture and nutritional properties and are consumed raw, roasted or processed into cakes, confectionery, ice cream, salads and other food products. Macadamia is represented by several species in Australia, but *M. integrifolia*, *M. tetraphylla* and their hybrids are predominantly cultivated commercially. Both species possess a somatic chromosome number of $2n = 28$. *M. integrifolia* is commonly known as the smooth-shelled type, producing high-quality kernels, whereas *M. tetraphylla* is characterised by a rough, pebbled shell and slightly elongated nuts.

Macadamia is primarily propagated through seeds for raising rootstocks, whereas vegetative propagation through grafting is preferred for the multiplication of superior cultivars and early bearing. Different grafting techniques, including simple, whip, side, wedge and cleft grafting are used for macadamia propagation. Well-matured, healthy nuts are commonly used for raising rootstock seedlings (Tripathi *et al.*, 2023) [2]. Suitable rootstock seedlings are generally selected for grafting, based on their age and stem thickness. Grafted plants have the advantage of maintaining the desirable characteristics of the scion and generally attain bearing earlier than seedling plants. Therefore, efficient grafting techniques are essential for rapid multiplication and establishment of elite macadamia planting material.

Graft union formation is a complex physiological process involving wound healing, callus formation, cambial reconnection and subsequent development of vascular tissues. Plant growth regulators, particularly auxins and cytokinin, play important roles in cell division, differentiation and vascular development during graft union formation. Following grafting, auxin accumulation in the scion and cytokinin activity in the rootstock contribute to wound responses and vascular tissue differentiation at the graft interface (Melnyk *et al.*, 2015) [3]. Hence, the use of suitable growth regulators may enhance callus formation, improve vascular connection

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and increase grafting success. Hence, the present study has been conducted with the objectives to study the effect of bio-regulators on grafting success and graft survivability of *Macadamia* nut.

Material and Methods

The study was conducted from November 2025 to June 2026 at Varashree Nursery, Gajanuru, Shivamogga district, *Karnataka*. The site lies in the Southern Transitional Zone (Zone 7) of *Karnataka* at an altitude of 570-588 m above mean sea level. The mean maximum temperature during the experimental period was 32.32 °C, the mean minimum temperature was 19.28 °C and relative humidity ranged from 32.50 to 91.50 per cent. The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications. There were 14 treatments, 20 grafts per treatment and 840 grafts in total were taken for the experiment. The treatments comprised of viz., T₀ - Control (No dip), T₁ - Water dip, T₂ - NAA @ 10 ppm, T₃ - NAA @ 20 ppm, T₄ - NAA @ 30 ppm, T₅ - BAP @ 10 ppm, T₆ - BAP @ 20 ppm, T₇ - BAP @ 30 ppm, T₈ - GA₃ @ 5 ppm, T₉ - GA₃ @ 10 ppm, T₁₀ - GA₃ @ 15 ppm, T₁₁ - Biovita @ 2500 ppm, T₁₂ - Biovita @ 5000 ppm and T₁₃ - Biovita @ 7500 ppm. Grafting was carried out during November-December using 3-4 months old *M. integrifolia* scions and 5-6 months old *M. tetraphylla* rootstocks were used for grafting with the following treatment details.

The one-season-old, pencil-thick, healthy scion shoots were selected from regular-bearing mother plants. Leaves were removed while retaining the petiole and curing was carried out ten days before grafting. Scions were collected during the morning, wrapped in moist cloth and grafted on the same day after treatment with 0.1 per cent Bavistin. Wedge grafting was followed, the scion was dipped in the respective

bio-regulator solution and inserted into the V-shaped cleft of the root-stock. The graft union was secured with polythene strip and grafts were maintained under polyhouse conditions.

Observations were recorded at monthly intervals at 60, 90, 120 and 150 days after grafting (DAG), with sprouting percentage additionally recorded at 30 and 45 DAG. Days to sprouting, sprouting percentage, graft success, scion girth, scion length and graft survivability were considered. Fisher's analysis of variance was used following Panse *et al.* (1967) [4] and critical difference (CD) values were worked out at *P* = 0.05 wherever the *F*-test was significant.

Results and Discussion

The bio-regulator treatments significantly influenced the selected grafting and growth parameters. Results and their interpretation are presented together for each parameter.

Among all the different bio-regulator treatments evaluated, the scion treated with (T₉) GA₃ @ 10 ppm recorded the earliest sprouting (22.86 days), followed by T₈ (24.13 days) and T₁₀ (24.14 days), whereas T₀ required the longest period (36.47 days) (Table 1.). Sprouting percentage was highest in T₄ (NAA @ 30 ppm) at 30, 45 and 60 DAG (60.84, 63.46 and 65.84%, respectively), while T₀ recorded 0.00, 27.15 and 31.82 per cent. The earlier sprouting under GA₃ may be associated with its role in promoting shoot elongation and active growth. Castro *et al.* (1991) [5] reported growth-promoting effects of GA₃ in macadamia rootstocks. The higher sprouting percentage under NAA may be related to auxin-mediated callus development and vascular reconnection at the graft interface processes, important for graft establishment (Pina *et al.*, 2005) [6].

Table 1. Effect of bio-regulators on days taken for sprouting and sprouting per cent at different intervals after grafting in macadamia nut

Treatments	Days taken for sprouting	Sprouting percent at different intervals		
		30 DAG	45 DAG	60 DAG
T ₀ : Control (No dip)	36.47 ^a	0.00 ^j	27.15 ^k	31.82 ^j
T ₁ : Water dip	31.51 ^b	31.25 ^h	37.18 ^h	39.86 ^g
T ₂ : NAA @ 10 ppm	26.60 ^d	39.86 ^d	55.82 ^c	57.74 ^c
T ₃ : NAA @ 20 ppm	25.46 ^e	43.62 ^b	58.34 ^b	60.68 ^b
T ₄ : NAA @ 30 ppm	24.42 ^f	60.84 ^a	63.46 ^a	65.84 ^a
T ₅ : BAP @ 10 ppm	27.34 ^d	33.58 ^g	35.74 ⁱ	38.52 ^h
T ₆ : BAP @ 20 ppm	26.82 ^d	35.42 ^f	39.24 ^g	41.06 ^f
T ₇ : BAP @ 30 ppm	28.22 ^c	31.04 ^{hi}	37.86 ^h	39.98 ^g
T ₈ : GA ₃ @ 5 ppm	24.13 ^f	38.64 ^e	46.88 ^f	49.36 ^e
T ₉ : GA ₃ @ 10 ppm	22.86 ^g	41.82 ^c	50.64 ^d	52.46 ^d
T ₁₀ : GA ₃ @ 15 ppm	24.14 ^f	42.34 ^c	47.92 ^e	50.18 ^e
T ₁₁ : Biovita @ 2500 ppm	26.81 ^d	34.86 ^f	37.84 ^h	40.12 ^{fg}
T ₁₂ : Biovita @ 5000 ppm	28.22 ^c	30.18 ⁱ	35.12 ^{ij}	36.94 ⁱ
T ₁₃ : Biovita @ 7500 ppm	28.22 ^c	33.04 ^g	34.56 ^j	37.62 ^{hi}
S. Em. ±	0.21	0.29	0.32	0.33
CD @ 5%	0.62	0.84	0.94	0.96

DAG: Days after grafting

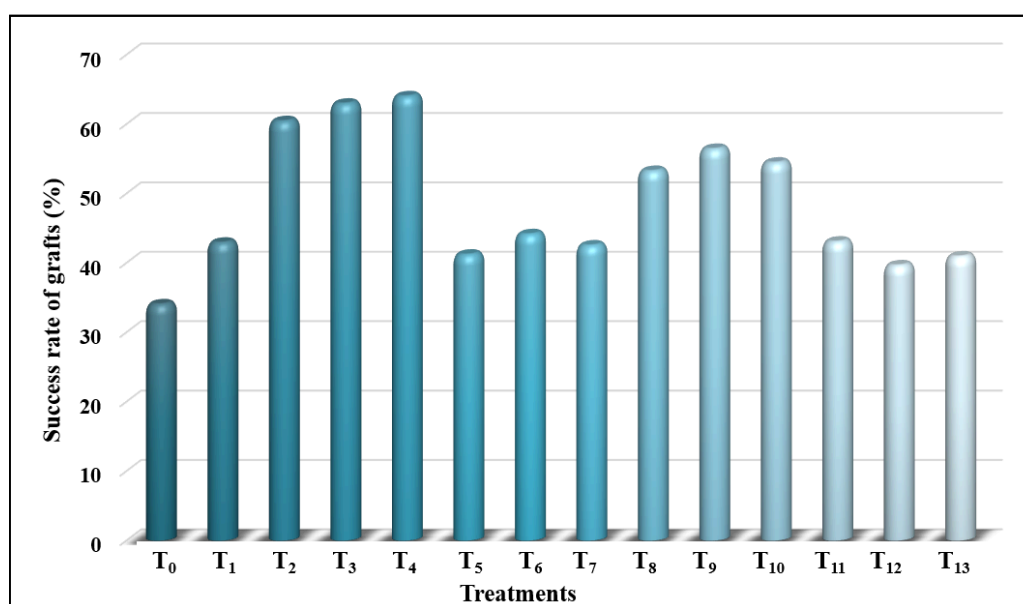
Significant differences were observed among the bio-regulator treatments for graft success at different stages after grafting (Table 2. & Fig. 1.). At 60 days after grafting (DAG), T₄ (NAA @ 30 ppm) recorded the highest graft success (61.84%), followed by T₃ (NAA @ 20 ppm) (58.42%), whereas the control (T₀) recorded the lowest (30.48%). At 90 DAG, T₄ recorded the maximum success rate (61.84%), followed by T₃ (58.42%), while T₀ recorded the minimum (32.64%). At 120 DAG, the highest graft success was recorded in T₄ (64.22%), followed by T₃ (63.16%), whereas T₀ recorded the lowest

(34.22%). At 150 DAG, T₄ maintained the highest graft success (64.04%), followed by T₃ (62.98%), while T₀ recorded the minimum (34.06%). The superior performance of NAA may be attributed to enhanced cell division, callus proliferation, cambial activity and vascular tissue differentiation at the graft interface. Similar findings were reported by Sharma *et al.* (2019) [7], Castro *et al.* (1991) [5], Choudhury *et al.* (2021) [8], Aminzadeh *et al.* (2013) [9], Ahmed *et al.* (2012) [10] and Pina *et al.* (2005) [6].

Table 2. Effect of bio-regulators on success rate of graft at different intervals after grafting in macadamia nut

Treatments	Success rate of graft (%) at different intervals			
	60 DAG	90 DAG	120 DAG	150 DAG
T ₀ : Control (No dip)	30.48 ^j	32.64 ^j	34.22 ^k	34.06 ^k
T ₁ : Water dip	38.68 ^g	38.68 ^g	43.06 ^h	42.94 ^h
T ₂ : NAA @ 10 ppm	55.86 ^c	55.86 ^c	60.64 ^c	60.46 ^c
T ₃ : NAA @ 20 ppm	58.42 ^b	58.42 ^b	63.16 ^b	62.98 ^b
T ₄ : NAA @ 30 ppm	61.84 ^a	61.84 ^a	64.22 ^a	64.04 ^a
T ₅ : BAP @ 10 ppm	37.18 ^h	37.18 ^h	41.38 ⁱ	41.22 ⁱ
T ₆ : BAP @ 20 ppm	39.68 ^f	39.68 ^f	44.16 ^g	44.16 ^g
T ₇ : BAP @ 30 ppm	38.52 ^g	38.52 ^g	42.72 ^h	42.54 ^h
T ₈ : GA ₃ @ 5 ppm	47.84 ^e	47.84 ^e	53.46 ^f	53.28 ^f
T ₉ : GA ₃ @ 10 ppm	50.96 ^d	50.96 ^d	56.68 ^d	56.46 ^d
T ₁₀ : GA ₃ @ 15 ppm	48.76 ^e	48.76 ^e	54.48 ^e	54.48 ^e
T ₁₁ : Biovita @ 2500 ppm	38.86 ^{fg}	38.86 ^{fg}	43.28 ^{gh}	43.10 ^h
T ₁₂ : Biovita @ 5000 ppm	35.84 ⁱ	35.84 ⁱ	39.86 ^j	39.68 ^j
T ₁₃ : Biovita @ 7500 ppm	36.56 ^{hi}	36.56 ^{hi}	40.96 ⁱ	40.96 ⁱ
S. Em. ±	0.32	0.33	0.34	0.34
CD @ 5%	0.94	0.97	0.99	0.99

DAG: Days after grafting

**Figure 1.** Effect of bio-regulators on success rate of grafts of Macadamia nut

Significant differences were observed among the bio-regulator treatments for scion girth throughout the observation period (Table 3.). Before grafting, scion girth ranged from 4.08 to 4.31 mm. At 60 days after grafting (DAG), T₄ (NAA @ 30 ppm) recorded the maximum scion girth (8.18 mm), followed by T₃ (NAA @ 20 ppm) (6.10 mm), whereas the minimum was recorded in T₀ (Control) (4.89 mm). At 90 DAG, T₄ recorded the highest value (8.31 mm), followed by T₃ (7.62 mm), while T₀ recorded the lowest (5.06 mm). At 120 DAG, T₄

recorded the maximum scion girth (8.96 mm), followed by T₃ (7.52 mm), whereas T₀ recorded the minimum (5.19 mm). At 150 DAG, T₄ recorded the highest scion girth (9.10 mm), followed by T₃ (8.84 mm), while T₀ recorded the lowest (5.51 mm). The enhanced girth of scion under NAA may be attributed to increased cambial activity, callus proliferation and vascular differentiation. Similar findings were reported by Choudhury *et al.* (2021) [8], Castro *et al.* (1991) [5], Hartmann *et al.* (2018) [11] and Ahmed *et al.* (2012) [10].

Table 3. Effect of bio-regulators on scion girth at different intervals after grafting in macadamia nut

Treatments	Scion girth (mm) at different intervals				
	Before grafting	60 DAG	90 DAG	120 DAG	150 DAG
T ₀ : Control (No dip)	4.12	4.89 ^e	5.06 ⁱ	5.19 ^g	5.51 ^e
T ₁ : Water dip	4.08	4.92 ^e	5.28 ^{hi}	5.62 ^g	6.73 ^d
T ₂ : NAA @ 10 ppm	4.25	5.87 ^{bc}	6.60 ^c	7.34 ^{bcd}	8.11 ^b
T ₃ : NAA @ 20 ppm	4.28	6.10 ^b	7.62 ^b	7.52 ^b	8.84 ^a
T ₄ : NAA @ 30 ppm	4.31	8.18 ^a	8.31 ^a	8.96 ^a	9.10 ^a
T ₅ : BAP @ 10 ppm	4.15	5.34 ^{cde}	6.12 ^{cdef}	7.11 ^{bcde}	7.38 ^c
T ₆ : BAP @ 20 ppm	4.10	5.87 ^{bc}	6.71 ^c	7.46 ^{bc}	8.21 ^b
T ₇ : BAP @ 30 ppm	4.22	4.99 ^e	5.75 ^{fgh}	6.61 ^{ef}	7.17 ^{cd}
T ₈ : GA ₃ @ 5 ppm	4.18	5.42 ^{cde}	6.23 ^{cdef}	7.15 ^{bcde}	7.46 ^c
T ₉ : GA ₃ @ 10 ppm	4.26	5.42 ^{cde}	6.38 ^{cde}	7.16 ^{bcde}	7.66 ^{bc}
T ₁₀ : GA ₃ @ 15 ppm	4.20	5.68 ^{bcd}	6.55 ^{cd}	7.26 ^{bcd}	8.08 ^b
T ₁₁ : Biovita @ 2500 ppm	4.14	5.27 ^{cde}	5.97 ^{def}	6.83 ^{cdef}	7.35 ^c
T ₁₂ : Biovita @ 5000 ppm	4.30	4.95 ^e	5.36 ^{ghi}	6.31 ^f	7.07 ^{cd}
T ₁₃ : Biovita @ 7500 ppm	4.16	5.23 ^{de}	5.90 ^{efg}	6.78 ^{def}	7.27 ^{cd}
S. Em. ±	0.09	0.11	0.10	0.11	0.11
CD @ 5%	NS	0.31	0.31	0.32	0.33

DAG: Days after grafting

Significant differences were observed among the bio-regulator treatments for scion length at different stages after grafting. Scion length increased progressively with advancement in the observation period (Table 4.). At 60, 90, 120 and 150 days after grafting (DAG), T₉ (GA₃ @ 10 ppm) recorded the maximum scion length of 19.58, 28.30, 34.50 and 37.68 cm, respectively, whereas the control (T₀) recorded the minimum values of 9.47, 12.51, 18.23 and 20.28 cm, respectively.

The enhanced scion elongation under GA₃ may be attributed to stimulation of cell division and elongation, rapid extension of internodal tissues and improved translocation of assimilates towards actively growing tissues. The present findings are in accordance with Castro *et al.* (1991) [5], Choudhury *et al.* (2021) [8], Aminzadeh *et al.* (2013) [9] and Ahmed *et al.* (2012) [10], who reported improved shoot growth following growth-regulator application.

Table 4. Effect of bio-regulators on scion length at different intervals after grafting in macadamia nut

Treatments	Scion length (cm) at different intervals				
	Before grafting	60 DAG	90 DAG	120 DAG	150 DAG
T ₀ : Control (No dip)	7.12	9.47 ⁱ	12.51 ^h	18.23 ^g	20.28 ⁱ
T ₁ : Water dip	7.08	10.02 ^{hi}	15.18 ^g	25.95 ^f	28.73 ^h
T ₂ : NAA @ 10 ppm	7.25	12.23 ^e	18.42 ^{def}	31.47 ^{cd}	33.93 ^{de}
T ₃ : NAA @ 20 ppm	7.31	13.37 ^{cd}	18.65 ^{cde}	32.35 ^{bc}	34.69 ^{cd}
T ₄ : NAA @ 30 ppm	7.28	13.37 ^{cd}	19.22 ^{bcd}	32.44 ^b	35.04 ^{bc}
T ₅ : BAP @ 10 ppm	7.15	12.23 ^e	18.19 ^{ef}	31.24 ^d	33.82 ^{de}
T ₆ : BAP @ 20 ppm	7.10	12.98 ^d	18.42 ^{def}	32.28 ^{bc}	34.63 ^{cd}
T ₇ : BAP @ 30 ppm	7.22	11.22 ^{fg}	17.54 ^f	26.81 ^f	30.16 ^g
T ₈ : GA ₃ @ 5 ppm	7.18	13.77 ^c	19.34 ^{bc}	32.54 ^b	35.60 ^b
T ₉ : GA ₃ @ 10 ppm	7.26	19.58 ^a	28.30 ^a	34.50 ^a	37.68 ^a
T ₁₀ : GA ₃ @ 15 ppm	7.20	15.08 ^b	19.58 ^b	33.98 ^a	35.65 ^b
T ₁₁ : Biovita @ 2500 ppm	7.14	11.89 ^{ef}	18.09 ^{ef}	30.85 ^{de}	33.40 ^{ef}
T ₁₂ : Biovita @ 5000 ppm	7.30	10.59 ^{gh}	17.50 ^f	26.63 ^f	29.07 ^h
T ₁₃ : Biovita @ 7500 ppm	7.16	11.54 ^{ef}	17.98 ^{ef}	30.00 ^e	32.60 ^f
S. Em. ±	0.12	0.17	0.24	0.26	0.26
CD @ 5%	NS	0.50	0.70	0.77	0.77

DAG: Days after grafting

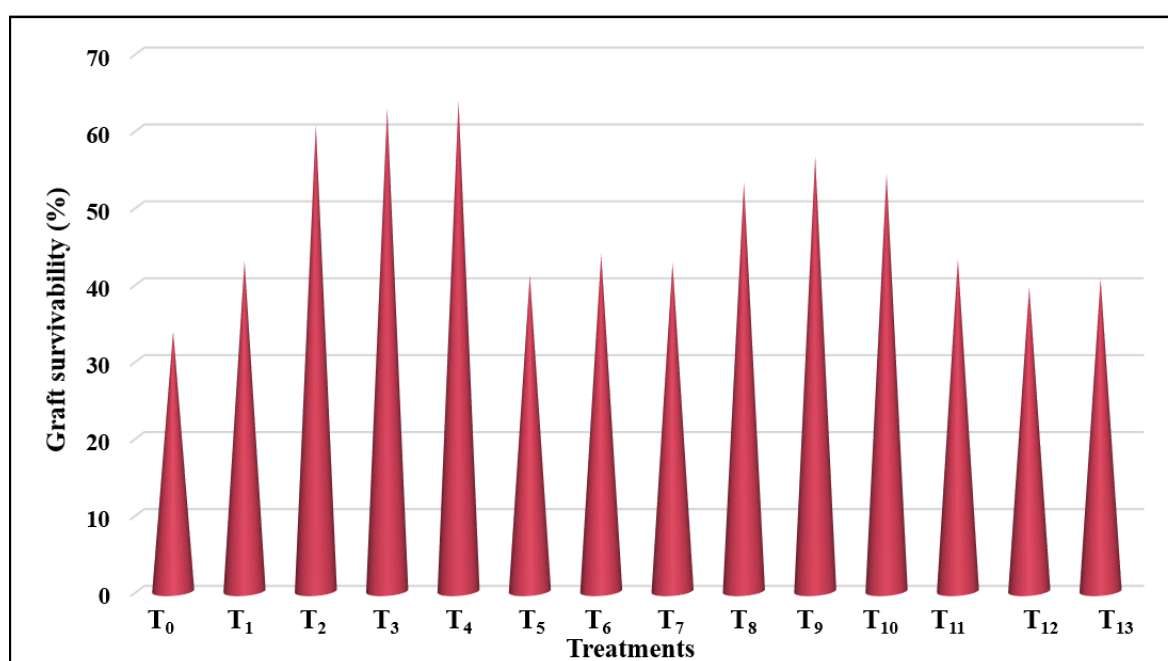
Graft survivability was significantly influenced by the different bio-regulator treatments at 120 and 150 days after grafting (DAG) (Table 5. & Fig. 2.). At 120 DAG, the highest graft survivability was recorded in T₄ – NAA @ 30 ppm (64.02%), followed by T₃ – NAA @ 20 ppm (63.02%), while the lowest was observed in T₀ (Control -34.08%). At 150 DAG, T₄ recorded the highest graft survivability (63.78%), which was statistically on par with T₃ (62.84%), whereas T₀ recorded the lowest value (33.82%). The enhanced survivability under NAA treatment might be attributed to auxin-mediated tissue re-

generation, cambial continuity and vascular reconnection, resulting in stronger graft union formation. Improved vascular continuity facilitates efficient movement of water and nutrients between rootstock and scion, thereby reducing post-grafting stress. The findings are in close agreement with Sharma *et al.* (2019) [7], Pina *et al.* (2005) [6] and Ahmed *et al.* (2012) [10], who highlighted the importance of auxins, callus formation and vascular reconnection in successful graft establishment.

Table 5. Effect of bio-regulators on graft survivability at different intervals after grafting in macadamia nut

Treatments	Graft survivability (%)	
	120 DAG	150 DAG
T ₀ : Control (No dip)	34.08 ^k	33.82 ^j
T ₁ : Water dip	42.96 ^h	42.96 ^{fg}
T ₂ : NAA @ 10 ppm	60.64 ^c	60.64 ^b
T ₃ : NAA @ 20 ppm	63.02 ^b	62.84 ^a
T ₄ : NAA @ 30 ppm	64.02 ^a	63.78 ^a
T ₅ : BAP @ 10 ppm	41.32 ⁱ	41.08 ^h
T ₆ : BAP @ 20 ppm	44.06 ^g	43.84 ^f
T ₇ : BAP @ 30 ppm	42.68 ^h	42.68 ^g
T ₈ : GA ₃ @ 5 ppm	53.34 ^f	53.12 ^e
T ₉ : GA ₃ @ 10 ppm	56.52 ^d	56.52 ^c
T ₁₀ : GA ₃ @ 15 ppm	54.42 ^e	54.24 ^d
T ₁₁ : Biovita @ 2500 ppm	43.18 ^{gh}	43.18 ^{fg}
T ₁₂ : Biovita @ 5000 ppm	39.82 ^j	39.56 ⁱ
T ₁₃ : Biovita @ 7500 ppm	40.84 ⁱ	40.62 ^h
S. Em. ±	0.34	0.34
CD @ 5%	0.99	0.99

DAG: Days after grafting

**Figure 2.** Effect of bio-regulators on graft survivability of Macadamia nut

Conclusion

The present investigation demonstrated that bio-regulators influenced graft establishment, scion growth and survivability in macadamia nut. NAA @ 30 ppm (T₄) was the most effective treatment for sprouting percentage, graft success, scion girth and graft survivability, whereas GA₃ @ 10 ppm (T₉) was most effective for reducing days taken for sprouting and increasing scion length. Therefore, NAA @ 30 ppm is promising for graft establishment and survivability, while GA₃ @ 10 ppm is promising for early sprouting and vigorous scion growth under the experimental conditions.

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