

Journal of Applied Biology & Biotechnology

Available online at: <https://jabonline.in/>

Optimization of conditions to achieve a high content of alizarin and purpurin in the adventitious roots of *Rubia cordifolia* L.

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DOI: <https://doi.org/10.7324/JABB.2024.194422>

SUPPLEMENTARY MATERIAL

Supplementary Table 1. Influence of various growth regulators and pH towards adventitious root formation from nodal explants of *R. cordifolia*.

Auxin type	Auxin concentration (mg/L)	pH	Biomass		Growth index	Total AQs (mg/g)	Alizarin (mg/g)	Purpurin (mg/g)
			Fresh weight (g/ 50 mL)	Dry weight (g/ 50 mL)				
Control	0	5.6	2.08 ± 0.04 ^w	0.64 ± 0.03 ^q	1.08 ± 0.04 ^w	6.00 ± 0.10 ^w	0.39 ± 0.06 ^{rs}	0.81 ± 0.04 ^r
		5.7	3.43 ± 0.78 ^v	0.84 ± 0.03 ^{opq}	2.43 ± 0.08 ^v	10.01 ± 0.10 ^u	0.57 ± 0.05 ^{qrs}	1.20 ± 0.06 ^p
		5.8	3.32 ± 0.39 ^v	0.68 ± 0.04 ^{pq}	2.32 ± 0.04 ^v	7.97 ± 0.10 ^v	0.34 ± 0.06 ^{rs}	0.88 ± 0.08 ^r

IBA	0.5	5.6	5.33 ± 0.68 ^u	1.05 ± 0.05 ^{nopq}	4.33 ± 0.07 ^u	11.90 ± 0.41 ^t	0.73 ± 0.06 ^{pqr}	1.23 ± 0.13 ^p
		5.7	6.65 ± 0.15 ^t	1.65 ± 0.04 ^{lmn}	5.64 ± 0.126 ^t	1.11 ± 0.07 ^x	1.56 ± 0.16 ^{kl}	0.84 ± 0.09 ^r
		5.8	6.26 ± 0.06 ^t	1.46 ± 0.05 ^{no}	5.26 ± 0.06 ^t	0.86 ± 0.09 ^x	3.33 ± 0.28 ^{de}	2.22 ± 0.12 ^{lm}
	1.0	5.6	8.71 ± 0.07 ^q	2.71 ± 0.09 ^{jk}	7.71 ± 0.07 ^q	15.99 ± 0.10 ^p	1.54 ± 0.16 ^{klm}	2.22 ± 0.12 ^{lm}
		5.7	11.53 ± 0.53 ⁿ	3.28 ± 0.11 ^{ij}	10.53 ± 0.05 ⁿ	19.69 ± 0.44 ^l	1.90 ± 0.09 ⁱ	2.45 ± 0.25 ^k
		5.8	11.28 ± 0.06 ⁿ	3.17 ± 0.06	10.28 ± 0.05 ⁿ	18.21 ± 0.19 ^{no}	1.66 ± 0.08 ^{ijk}	2.11 ± 0.11 ^m
	1.5	5.6	13.04 ± 0.09 ^m	3.33 ± 0.19 ⁱ	12.04 ± 0.09 ^m	19.93 ± 0.27 ^l	1.81 ± 0.13 ^{ij}	2.13 ± 0.09 ^m
		5.7	14.01 ± 0.08 ^k	6.08 ± 2.98 ^{abc}	13.01 ± 0.08 ^k	22.05 ± 0.19 ^h	2.46 ± 0.25 ^h	2.71 ± 0.33 ^{ij}
		5.8	13.57 ± 0.21 ^l	3.55 ± 0.33 ^{hi}	12.57 ± 0.21 ^l	21.03 ± 0.15 ^{ij}	1.88 ± 0.12 ⁱ	2.36 ± 0.15 ^{kl}
	2.0	5.6	14.11 ± 0.13 ^{jk}	4.21 ± 0.17 ^{fg}	13.11 ± 0.13 ^{jk}	19.16 ± 0.21 ^m	2.60 ± 0.19 ^{gh}	2.99 ± 0.11 ^{fgh}
		5.7	14.47 ± 0.11 ^{ijk}	4.46 ± 0.18 ^{fg}	13.47 ± 0.11 ^{ijk}	22.16 ± 0.20 ^h	2.57 ± 0.39 ^h	3.33 ± 0.52 ^e
		5.8	14.57 ± 0.07 ^{ij}	4.17 ± 0.07 ^{fgh}	13.57 ± 0.07 ^{ij}	20.05 ± 0.83 ^{kl}	2.54 ± 0.13 ^h	2.84 ± 0.13 ^{hi}
	2.5	5.6	14.44 ± 0.22 ^{ijk}	4.38 ± 0.10 ^{fg}	13.44 ± 0.22 ^{ijk}	19.23 ± 0.32 ^m	2.63 ± 0.21 ^{gh}	2.53 ± 0.06 ^{jk}
		5.7	15.31 ± 0.22 ^h	4.81 ± 0.12 ^{ef}	14.31 ± 0.22 ^h	24.99 ± 0.24 ^f	3.33 ± 0.28 ^{de}	3.16 ± 0.47 ^{efg}
		5.8	14.85 ± 0.38 ⁱ	4.29 ± 0.16 ^{fg}	13.85 ± 0.38 ⁱ	20.79 ± 0.80 ^j	2.94 ± 0.06 ^f	3.09 ± 0.10 ^{efg}
IAA	0.5	5.6	13.54 ± 0.41 ^l	4.07 ± 0.09 ^{gh}	12.54 ± 0.41 ^l	15.21 ± 0.27 ^q	2.46 ± 0.17 ^h	2.96 ± 0.05 ^{gh}
		5.7	14.05 ±	4.29 ±	13.05 ±	18.39 ±	2.51 ±	3.22 ±

			0.33 ^k	0.13 ^{fg}	0.33 ^k	0.47 ⁿ	0.37 ^h	0.12 ^{ef}
		5.8	14.34 ± 0.40 ^{jk}	4.35 ± 0.29 ^{fg}	13.34 ± 0.40 ^{jk}	16.15 ± 0.20 ^p	2.39 ± 0.24 ^h	3.16 ± 0.12 ^{efg}
	1.0	5.6	17.48 ± 0.47 ^g	4.35 ± 0.26 ^{fg}	16.48 ± 0.47 ^g	20.27 ± 0.42 ^k	2.51 ± 0.39 ^h	3.22 ± 0.06 ^{ef}
		5.7	19.27 ± 0.37 ^e	5.30 ± 0.14 ^{de}	18.27 ± 0.37 ^e	24.10 ± 0.11 ^g	3.39 ± 0.32 ^{de}	3.68 ± 0.08 ^d
		5.8	18.76 ± 0.26 ^f	4.64 ± 0.33 ^{fg}	17.76 ± 0.26 ^f	21.39 ± 0.39 ⁱ	2.56 ± 0.30 ^h	2.85 ± 0.41 ^{hi}
	1.5	5.6	21.92 ± 0.73 ^b	6.16 ± 0.13 ^{ab}	20.93 ± 0.73 ^a	26.89 ± 0.18 ^e	3.50 ± 0.29 ^{cde}	4.17 ± 0.10 ^b
		5.7	23.53 ± 0.95^a	6.41 ± 0.37^a	22.53 ± 0.95^a	32.00 ± 0.10^a	4.73 ± 0.13^a	4.63 ± 0.11^a
		5.8	21.12 ± 0.38 ^c	6.39 ± 0.25 ^a	20.12 ± 0.38 ^c	29.18 ± 0.28 ^c	4.29 ± 0.29 ^b	4.20 ± 0.11 ^b
	2.0	5.6	19.21 ± 0.30 ^e	5.61 ± 0.14 ^{bcd}	18.21 ± 0.30 ^e	28.01 ± 0.11 ^d	3.28 ± 0.25 ^e	3.64 ± 0.17 ^d
		5.7	20.59 ± 0.65 ^d	6.27 ± 0.27 ^a	19.59 ± 0.65 ^d	30.17 ± 0.19 ^b	3.62 ± 0.33 ^{cd}	4.07 ± 0.10 ^{bc}
		5.8	19.25 ± 0.41 ^e	5.46 ± 0.34 ^{cde}	18.25 ± 0.41 ^e	30.14 ± 0.25 ^b	3.73 ± 0.20 ^c	3.90 ± 0.11 ^c
	2.5	5.6	19.34 ± 0.33 ^e	5.30 ± 0.22 ^{de}	18.34 ± 0.34 ^e	29.22 ± 0.3 ^c	2.88 ± 0.18 ^{fg}	3.31 ± 0.22 ^e
		5.7	19.33 ± 0.59 ^e	5.39 ± 0.27 ^{de}	18.33 ± 0.59 ^e	30.37 ± 0.50 ^b	3.47 ± 0.28 ^{cde}	3.67 ± 0.15 ^d
		5.8	19.14 ± 0.21 ^{ef}	5.35 ± 0.17 ^{de}	18.14 ± 0.21 ^{ef}	28.91 ± 0.27 ^c	3.31 ± 0.16 ^e	3.06 ± 0.10 ^{fgh}
NAA	0.5	5.6	8.31 ± 0.23 ^{qr}	1.63 ± 0.44 ^{lmn}	7.31 ± 0.2325 ^{qr}	13.10 ± 0.20 ^s	0.71 ± 0.19 ^{pqr}	1.14 ± 0.07 ^p
		5.7	9.31 ± 0.20 ^p	2.40 ± 0.19 ^k	8.31 ± 0.20 ^p	15.15 ± 0.22 ^q	1.32 ± 0.19	1.86 ± 0.08 ⁿ
		5.8	9.61 ± 0.47 ^p	2.14 ± 0.06 ^{klm}	8.61 ± 0.47 ^p	14.06 ± 0.13 ^r	1.14 ± 0.19 ^{no}	0.90 ± 0.06 ^{qr}

	1.0	5.6	7.59 ± 0.39 ^s	1.28 ± 0.13 ^{nopq}	6.59 ± 0.39 ^s	20.22 ± 0.24 ^k	0.88 ± 0.07 ^{op}	1.24 ± 0.13 ^p
		5.7	8.38 ± 0.40 ^{qr}	2.15 ± 0.10 ^{klm}	7.38 ± 0.40 ^{qr}	22.26 ± 0.33 ^h	1.19 ± 0.14 ⁿ	1.53 ± 0.13 ^o
		5.8	8.20 ± 0.18 ^r	2.18 ± 0.14 ^{klm}	7.20 ± 0.18 ^r	19.00 ± 0.15 ^m	0.80 ± 0.05 ^{pqr}	1.11 ± 0.11 ^{pq}
	1.5	5.6	9.31 ± 0.35 ^p	2.31 ± 0.25 ^{klm}	8.31 ± 0.35 ^p	20.33 ± 0.33 ^k	1.31 ± 0.20 ^{lmn}	1.48 ± 0.12 ^o
		5.7	10.48 ± 0.42 ^o	3.12 ± 0.08 ^{ij}	9.48 ± 0.42 ^o	25.15 ± 0.35 ^f	1.40 ± 0.26 ^{klmn}	2.13 ± 0.09 ^m
		5.8	10.15 ± 0.04 ^o	2.48 ± 0.33 ^k	9.15 ± 0.04 ^o	19.27 ± 0.33 ^m	1.78 ± 0.16 ^{ij}	1.89 ± 0.07 ⁿ
	2.0	5.6	7.35 ± 0.36 ^s	1.23 ± 0.18 ^{nopq}	6.35 ± 0.36 ^s	16.10 ± 0.20 ^p	1.24 ± 0.10 ^{mn}	1.17 ± 0.08 ^p
		5.7	8.33 ± 0.37 ^{qr}	1.60 ± 0.37 ^{mn}	7.33 ± 0.37 ^{qr}	17.95 ± 0.17 ^o	1.32 ± 0.17 ^{lmn}	1.75 ± 0.08 ⁿ
		5.8	7.67 ± 0.33 ^s	1.35 ± 0.17 ^{nop}	6.67 ± 0.33 ^s	14.14 ± 0.16 ^r	1.10 ± 0.13 ^{no}	1.21 ± 0.08 ^p
	2.5	5.6	8.22 ± 0.22 ^r	2.22 ± 0.43 ^{klm}	7.22 ± 0.22 ^r	13.16 ± 0.20 ^s	0.68 ± 0.06 ^{pqr}	1.11 ± 0.07 ^{pq}
		5.7	9.37 ± 0.24 ^p	2.46 ± 0.32 ^k	8.37 ± 0.24 ^p	15.20 ± 0.20 ^q	0.87 ± 0.06 ^{opq}	1.89 ± 0.09 ⁿ
		5.8	8.25 ± 0.19 ^{qr}	2.15 ± 0.05 ^{klm}	7.25 ± 0.19 ^{qr}	12.13 ± 0.16 ^t	0.52 ± 0.04 ^{rs}	1.32 ± 0.11 ^{op}

* Means followed by different letters within a column are significantly different at $p \leq 0.05$.

Supplementary Fig. 1. Distribution map of sampling point in Kolli hills, Eastern Ghats, Tamil Nadu

