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Triacontanol enhances cytokinin-mediated *in vitro* shoot proliferation and biomass accumulation in two cherry rootstocks

Supplementary Material

Table S1. Chemical composition of full-strength MS basal medium (Murashige & Skoog, 1962)

Nutrient group type	Component	Concentration (mg L^{-1})
Macronutrients	Ammonium nitrate (NH_4NO_3)	1650
	Potassium nitrate (KNO_3)	1900
	Calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$)	440
	Magnesium sulfate heptahydrate ($MgSO_4 \cdot 7H_2O$)	370
	Potassium phosphate monobasic (KH_2PO_4)	170
Micronutrients	Boric acid (H_3BO_3)	6.2
	Cobalt chloride hexahydrate ($CoCl_2 \cdot 6H_2O$)	25
	Cupric sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$)	25
	Manganese sulfate monohydrate ($MnSO_4 \cdot H_2O$)	16.9
	Potassium iodide (KI)	0.83
	Molybdic acid (sodium salt) dihydrate ($Na_2MoO_4 \cdot 2H_2O$)	0.25
	Zinc sulfate heptahydrate ($ZnSO_4 \cdot 7H_2O$)	8.6
Iron Source (Chelated Iron)	Disodium EDTA ($Na_2EDTA \cdot 2H_2O$)	37.26
	Ferrous sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$)	27.8
Vitamins and Amino Acids	Myo-Inositol	100
	Nicotinic acid	0.5
	Pyridoxine · HCl	0.5
	Thiamine · HCl	0.1
	Glycine	2.0

Table S2. Main and interactive effects of TRIA and BA on *in vitro* shoot proliferation parameters in CAB-6P and Gisela 6 rootstocks (GLM / 2-way ANOVA)

Cherry rootstock	Treatments TRIA (μg L^{-1}) BA (mg L^{-1})	Shoot number/ explant	Shoot length (mm)	Shoot multiplication (%)	Shoot FW (g)	Shoot DW (g)	Shoot dry matter content (DW %)
CAB-6P	TRIA Concentration	0.000***	0.717 ns	0.000***	0.020*	0.027*	0.002**
	BA Concentration	0.000***	0.000***	0.000***	0.000***	0.000***	0.000***
	TRIA × BA	0.000***	0.224 ns	0.000***	0.001**	0.005**	0.113 ns
Gisela 6	TRIA Concentration	0.000***	0.367 ns	1.000 ns	0.000***	0.020*	0.000***
	BA Concentration	0.011*	0.000***	0.000***	0.611 ns	0.125 ns	0.014*
	TRIA × BA	0.023*	0.042*	1.000 ns	0.033*	0.083 ns	0.004**

ns: non-significant effect at the 5% level ($P > 0.05$); *, **, and ***: significant effect at the 5%, 1% and 0.1% level ($P \leq 0.05$, $P \leq 0.01$, and $P \leq 0.001$) respectively.

Table S3. Main and interactive effects of rootstock type, TRIA, and BA on *in vitro* shoot proliferation parameters in CAB-6P and Gisela 6 rootstocks (GLM / 3-way ANOVA)

Experimental factors	Shoot number/ explant	Shoot length (mm)	Shoot multiplication (%)	Shoot FW (g)	Shoot DW (g)	Shoot dry matter content (DW %)
Rootstock	0.000***	0.000***	0.000***	0.050*	0.122 ns	0.279 ns
TRIA	0.000***	0.886 ns	0.000***	0.000***	0.001**	0.013*
BA	0.000***	0.000***	0.000***	0.000***	0.000***	0.000***
Rootstock × TRIA	0.000***	0.220 ns	0.000***	0.000***	0.225 ns	0.000***
Rootstock × BA	0.000***	0.240 ns	0.000***	0.002**	0.436 ns	0.000***
TRIA × BA	0.015*	0.045*	0.000***	0.005**	0.041*	0.172 ns
Rootstock × TRIA × BA	0.000***	0.108 ns	0.000***	0.003**	0.017*	0.013*

ns: non-significant effect at the 5% level ($P > 0.05$); *, **, and ***: significant effect at the 5%, 1% and 0.1% level ($P \leq 0.05$, $P \leq 0.01$, and $P \leq 0.001$) respectively.

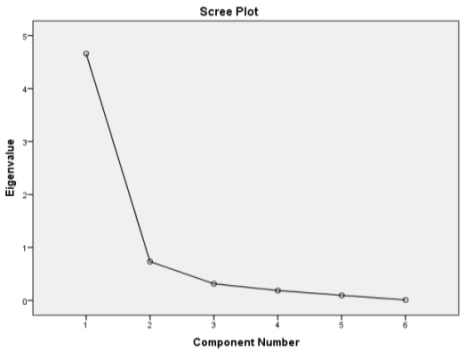
(a)

Communalities

	Initial	Extraction
Shoot_number	1.000	.863
Shoot_length	1.000	.905
Shoot_FW	1.000	.968
Shoot_DW	1.000	.943
Shoot_multiplication_percentage	1.000	.914
Shoot_dry_matter_content	1.000	.801

Extraction Method: Principal Component Analysis.

(b)



(c)

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	4.660	77.673	77.673	4.660	77.673	77.673	3.268	54.472	54.472
2	.733	12.224	89.896	.733	12.224	89.896	2.125	35.424	89.896
3	.314	5.231	95.128						
4	.188	3.127	98.255						
5	.096	1.598	99.852						
6	.009	.148	100.000						

Extraction Method: Principal Component Analysis.

(d)

Component Matrix[a]

	Component	
	1	2
Shoot_DW	.942	.238
Shoot_FW	.924	.338
Shoot_number	.898	.238
Shoot_dry_matter_content	-.893	
Shoot_multiplication_percentage	.854	-.429
Shoot_length	-.766	.565

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

(e)

Component Transformation Matrix

Component	1	2
1	.803	.595
2	.595	-.803

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

(f)

Rotated Component Matrix[a]

	Component	
	1	2
Shoot_FW	.943	.279
Shoot_DW	.898	.370
Shoot_number	.863	.344
Shoot_dry_matter_content	-.751	-.487
Shoot_length	-.279	-.910
Shoot_multiplication_percentage	.431	.853

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.[a]
a. Rotation converged in 3 iterations.

(g)

Component Score Coefficient Matrix

	Component	
	1	2
Shoot_number	.348	-.146
Shoot_length	.327	-.717
Shoot_FW	.433	-.252
Shoot_DW	.355	-.140
Shoot_multiplication_percentage	-.201	.579
Shoot_dry_matter_content	-.199	-.053

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.
Component Scores.

Fig. S1. PCA outputs for *in vitro* shoot proliferation traits in CAB-6P rootstock: (a) communalities; (b) scree plot; (c) total variance explained; (d) unrotated component matrix; (e) component transformation matrix; (f) rotated component matrix; (g) component score coefficient matrix

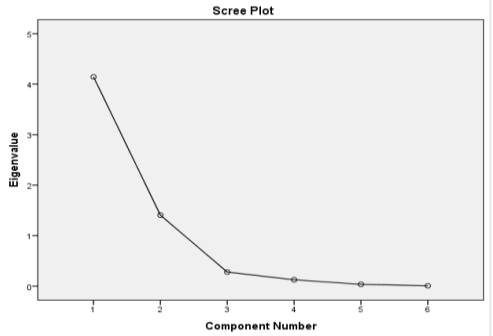
(a)

Communalities

	Initial	Extraction
Shoot_number	1.000	.958
Shoot_length	1.000	.945
Shoot_FW	1.000	.967
Shoot_DW	1.000	.845
Shoot_multiplication_percentage	1.000	.906
Shoot_dry_matter_content	1.000	.930

Extraction Method: Principal Component Analysis.

(b)



(c)

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings			Rotation Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	4.145	69.083	69.083	4.145	69.083	69.083	3.649	60.808	60.808
2	1.407	23.445	92.529	1.407	23.445	92.529	1.903	31.720	92.529
3	.279	4.648	97.177						
4	.127	2.109	99.286						
5	.036	.598	99.884						
6	.007	.116	100.000						

Extraction Method: Principal Component Analysis.

(d)

Component Matrix[a]

	Component	
	1	2
Shoot_dry_matter_content	-.964	
Shoot_number	.958	.200
Shoot_FW	.907	.380
Shoot_DW	.874	.285
Shoot_multiplication_percentage	.731	-.609
Shoot_length	-.419	.878

Extraction Method: Principal Component Analysis.
a. 2 components extracted.

(e)

Component Transformation Matrix

Component	1	2
1	.905	-.426
2	.426	.905

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.

(f)

Rotated Component Matrix[a]

	Component	
	1	2
Shoot_FW	.983	
Shoot_number	.952	-.227
Shoot_DW	.912	-.115
Shoot_dry_matter_content	-.884	.384
Shoot_length		.972
Shoot_multiplication_percentage	.403	-.862

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.[a]
a. Rotation converged in 3 iterations.

(g)

Component Score Coefficient Matrix

	Component	
	1	2
Shoot_number	.270	.030
Shoot_length	.174	.607
Shoot_FW	.313	.151
Shoot_DW	.277	.093
Shoot_multiplication_percentage	-.025	-.467
Shoot_dry_matter_content	-.219	.081

Extraction Method: Principal Component Analysis.
Rotation Method: Varimax with Kaiser Normalization.
Component Scores.

Fig. S2. PCA outputs for *in vitro* shoot proliferation traits in Gisela 6 rootstock: (a) communalities; (b) scree plot; (c) total variance explained; (d) unrotated component matrix; (e) component transformation matrix; (f) rotated component matrix; (g) component score coefficient matrix