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Influence of Growth regulators on Physiology and Senescence of Cut Stems of Chrysanthemum (Chrysanthemum morifolium Ramat) Var. Thai Ching Queen

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Abstract - The present study was conducted on cut stems of standard chrysanthemum var. Thai Ching Queen to assess the effect of ethrel, STS, 6-BAP and TDZ on post harvest senescence of florets and yellowing of leaves. The stems were harvested when $\frac{3}{4}$ th outer florets were fully expanded and treated with ethrel (50 and 100 μ M), STS (0.1 and 0.2 mM), 6-BAP (50 and 100 μ M) and TDZ (5 and 10 μ M) for 24 h. After the treatments, the stems were placed in distilled water. One set of stems were similarly treated with water represented control. The stems treated with ethrel exhibited early senescence of florets and yellowing of leaves which was associated with decrease in MSI, loss of chlorophyll and respiratory metabolites (total soluble and reducing sugars), accumulation of H₂O₂ content, increase in lipid peroxidation and loss in activity of antioxidant enzymes viz. peroxidase (POD), superoxide dismutase (SOD) and catalase (CAT). BAP and TDZ treatments prolonged post harvest longevity of florets as well as leaves. This effect was associated with increased activities of antioxidant enzymes. BAP and TDZ treatments, also reduced the production of H₂O₂, minimized lipid peroxidation, maintained respirable substrates and improved water uptake of the stems. TDZ was found to be most effective for delaying floret senescence as well as yellowing of leaves in cut stems of chrysanthemum.

Key words: Chrysanthemum, ethrel, floret senescence, leaf yellowing, pulsing, thidiazuron, vase-life.

I. INTRODUCTION

Maintaining good quality of cut flowers and extending the vase life are considered vital and practical for having acceptable products for the market. The three main factors which affect vase life of cut flowers are plant hormone ethylene which accelerates the senescence of many flowers, depletion of sugars and microorganisms which cause vascular blockage and thus reduce the vase life of cut flowers (Zencirkiran 2010). Numerous studies have been undertaken with the objective to evaluate treatments with various chemicals to extend vase life of various cut flower species (Redman et al. 2002; Macnish et al. 2008; Zencirkiran 2010; Mashhadian et al. 2012).

Chrysanthemum is ranked as the second most economically important cut flower in the world, after rose (Kafi and Ghahsareh 2009). The cut stems of chrysanthemum are highly prone to loss of water through stomata and cavitation of xylem vessels (Meeteren 1989; Nowak and Rudnicki 1990; Singh and Moore 1992). The stems of chrysanthemum varieties also exhibit pre-mature yellowing of leaves which adversely affects vase life. Therefore, finding methods to increase longevity of the cut stem is of great importance. Using preservatives in vase solutions is one of the most common methods for prolonging cut flowers' vase life. Physiology of flower senescence has predominantly been studied in some plant families such as Carophyllacaeae, Malvaceae and Orchidaceae in which petal senescence is regulated by ethylene (Trobacher 2009, Shahri and Tahir; 2011). The evolution of ethylene was not observed in chrysanthemum (Singh and Moore, 1992). Relatively few studies have been made on flower senescence in ethylene-insensitive flowers. The mechanism of floret and leaf senescence in chrysanthemum is not fully understood. The present study was aimed at the physiological and biochemical basis of petal and leaf senescence in chrysanthemum.

II. MATERIALS AND METHODS

Plants of standard chrysanthemum var. Thai Ching Queen were raised through the rooting of terminal cuttings. The rooted cuttings were planted 30×30 cm apart on 6 inch raised beds, at the Research Farm of the Department of Floriculture and Landscaping, PAU, Ludhiana. Recommended agronomical practices were followed to raise the plants to flowering stage. The stems were harvested when ³/₄th outer florets were fully expanded. Leaves from lower one-third portion of the stem were removed and uniform number of leaves were retained on each stem. The stems were pre-treated with 1000 mg l⁻¹ silver nitrate solution by dipping basal 2-3 cm portion for one min to prevent microbial growth on the cut surface of the stem. Thereafter, the stems were treated with the solution containing ethrel (50 and 100 µM), silver thiosulphate (STS) (0.1 and 0.2 mM), 6-benzylaminopurine (BAP) (50 and 100 μ M) and thidiazuron (TDZ) (5 and 10 µM), for 24 h in an air-conditioned laboratory (22±3°C; 60-70% R.H. under continuous illumination of 18 μ mol m⁻² s⁻¹ intensity provided by 40 W fluorescent tubes). Following the treatments, the stems were placed in distilled water. Keeping quality of the stems was evaluated under laboratory conditions mentioned above. The freshly-harvested stems placed in distilled water served as control. The experiment was set up in randomized block design with three replications and three stems in each replication. Observations were recorded for days to the initiation and complete senescence of florets and 50% yellowing of leaves. The amount of water uptake by stems was calculated by measuring the volume of water at the termination of vase life and subtracting it from the initial quantity of distilled water in jars. For biochemical estimations, samples of the florets were taken from outer, middle as well as inner whorls and pooled together where as leaves were collected from middle portion of the stem after 6 and 12 days in vase. Biochemical estimations were carried out for leaf chlorophyll content (Hiscox and Israelstam 1979), membrane stability index (Singh et al. 2007), total soluble sugars (Dubois et al. 1956), reducing sugars (Nelson 1944), H₂O₂ content (Brenan and Frenkel 1977), lipid peroxidation (Heath and Packer 1968) and activities of antioxidant enzymes viz. peroxidase (POD) (Shannon et al. 1966), superoxide dismutase (SOD) (Marklund and Marklund 1974) and catalase (CAT) (Teranishi et al. 1974).

Statistical analysis: Each value represents the mean of three independent replicates. The data has been analyzed statistically and CD computed at p=0.05 using CPCS1 software.

III. RESULTS AND DISCUSSION

Floret senescence and yellowing of leaves: Results presented in Table I show that the initiation of floret senescence occurred earliest of all in stems treated with 100 μ M ethrel, followed by those treated with 50 μ M of ethrel which indicates that ethylene plays role in inducing floret senescence in chrysanthemum. Silver thiosulphate (STS) at 0.1 and 0.2 mM concentration did not affect days to the initiation or complete senescence of florets. STS is an inhibitor of ethylene action and is reported to

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improve vase life of ethylene-sensitive flowers such as sweet pea (Mor et al. 1984), some varieties of rose (Chamani 2006), carnation (Karimi et al. 2013) etc. Singh and Moore (1992) have earlier reported that the flowers of chrysanthemum cv. Snow Don produced minute quantities of ethylene and exogenouslyapplied ethylene also did not affect floret longevity. Ethrel-induced floret senescence in var. Thai Ching Queen indicates that ethylene induces floret senescence of some chrysanthemum cultivars. . Pardha Saradi and Mohan Ram (1989) reported that cobalt chloride, an inhibitor of ethylene biosynthesis extended vase life of chrysanthemum in cv. Jyotsna. IAA, which enhances the activity of ACC synthase (Yang 1980), antagonized the effect of cobalt chloride which indicates that ethylene might induce senescence of florets in some cultivars of chrysanthemum. STS did not significantly affect days to the initiation or complete senescence of florets. It could be because STS treatment was below the threshold level required to delay senescence of florets. Both BAP and TDZ delayed the initiation as well as complete senescence of florets. TDZ was more effective than BAP in this respect. Ethrel treatment significantly hastened the yellowing of leaves. Yellowing of leaves was initiated in 4.00 and 3.33 days in stems treated with ethrel, 50 and 100 µM as compared to 8.67 days in control. Reyes-Arribas et al. (2000) have also reported that exogenous ethylene accelerated decline in chlorophyll content in detached leaves of 'Tara', a yellowing cultivar of chrysanthemum. Doi et al. (2004) also reported that ethylene promoted leaf yellowing in chrysanthemum cultivar 'Shuho-no-chikara' and the sensitivity of the leaves to ethylene gradually increased with time. Both BAP and TDZ delayed the appearance of symptoms of yellowing in the leaves. TDZ was more effective in this respect. The stems treated with TDZ at 5 and 10 µM concentrations exhibited the initiation of leaf senescence in 14.33 and 14.78 days respectively, as compared to 8.67 days in control. Similar trends were observed on the effect of treatments on 50% yellowing of leaves which occurred at the earliest in 100 µM ethrel-treated stems. Maximum delay in 50% yellowing of leaves was observed in stems treated with 10 µM of TDZ followed by the treatment with 5 µM TDZ. Some cut flowers such as stock, Lilium, Alstromeria and chrysanthemum are sensitive to post harvest yellowing of leaves (Ferrante et al. 2012). One of the main factors causing leaf yellowing could be ascribed to the degradation of chlorophyll caused by lack of cytokinins. BAP has been reported to extend vase life of 3 species of Draceana viz. D. marginata 'bicolor', D. sanderiana 'white' and D. deremensis whereas STS had no effect (Subhashini et al. 2011). Thidiazuron (TDZ), a substituted phenyl urea with cytokinin activity is reported to delay leaf yellowing as well as flower senescence in freesia, geranium, Ornithogalum, Euphorbia fulgens (Jiang et al. 2009), cut stock flowers and Matthiola incana (Ferrante et al. 2012).

Water absorption: Stems treated with ethrel (50 and 100 μ M) recorded the minimum water absorption. Ethrel-induced decrease in water absorption was apparently due to early senescence of leaves. Treatments with STS did not significantly affect water absorption by the stem over the control. BAP and TDZ improved water absorption and the effects were more pronounced in case of TDZ-treated stems. Improved water absorption in BAP and TDZ-treated stems was apparently due to delayed senescence of leaves and florets with these treatments. It has also been reported that treatment with TDZ in chrysanthemum prevented weight loss and improved hydraulic conductance of the stem (Ferrante 2012). It may be noted that in the present study though BAP and TDZ both delayed senescence of florets and leaves, TDZ was more effective at much lower concentrations i.e. at 5 and 10 μ M than BAP at 50 and 100 μ M. Jiang et al. (2009) have suggested that TDZ is a good substitute for BAP, zeatin and other cytokinins because of its high activity and non metabolizable nature.

Chlorophyll content: Chlorophyll content showed decline with increase in duration in vase (Figure I). The minimum chlorophyll content was observed in treatment with 100 μ M ethrel after 12 days in vase. STS treatment did not significantly affect chlorophyll content of the leaves. On the other hand, both BAP and TDZ maintained significantly higher chlorophyll content than the control. Effect of TDZ in this respect was more pronounced than that of BAP. After 12 days in vase, the maximum chlorophyll content was observed in leaves of stems treated with10 μ M TDZ. These results support the observations discussed earlier that TDZ treatment delayed number of days taken for yellowing of leaves.

Membrane stability index (MSI): Cut stems showed progressive decline in MSI in both florets and leaves with increase in number of days in vase (Figure II). The maximum MSI was observed on 0 day i.e. immediately after the treatment but declined to the lowest level after 12 days in vase. Treatment with ethrel significantly decreased where as BAP and TDZ substantially improved MSI of both florets and the leaves. MSI is an indicator of functional properties of the membrane. Membrane deterioration is characteristic feature of petal senescence in which an increase in membrane fluidity is observed and membranes lose selective permeability (Singh et al. 2007). Increase in membrane permeability and electrolyte leakage has also been reported earlier in senescing petals (Elanchezian and Srivastava 2001, Gulzar et al. 2005, Chakrabarty et al. 2009). Membrane stabilizing effect of BAP and TDZ indicates that both these bioregulators curtailed the deteriorative processes that bring about loss of membrane permeability in florets and leaves.

Total soluble sugar and reducing sugar content: Total soluble sugar and reducing sugar content of florets showed decline as number of days progressed in vase (Table II). Leaves recorded significantly lower total soluble as well as reducing sugar contents than florets. Florets from the stems treated with ethrel exhibited significant decrease in total soluble sugar content. The stems treated with both the concentrations of STS did not significantly influence total soluble sugar content over the control. Treatment with BAP and TDZ slowed down the reduction in total soluble sugar content as only slight decrease was observed after 6 and 12 days with these treatments. It was observed that BAP and TDZ treatments maintained high total soluble sugar contents even after 12 days. Exogenous ethylene treatment is reported to increase activities of hydrolytic enzymes which lead to rapid depletion of sugars and other metabolites consequently negatively affecting vase life of cut flowers (van Doorn and Woltering 2008). In the present study, STS treated stems did not exhibit significant change in total soluble and reducing sugar contents over the control. These treatments also did not prolong longevity of the florets or leaves. Cytokinins are reported to maintain high carbohydrate levels by regulating assimilate partitioning (Roitsch and Ehness 2000). Cytokinins also affect invertase activity responsible for sugar metabolism in florets and leaves (Lara et al. 2004). This may explain the maintenance of high soluble sugars of florets and leaves in stems treated with BAP and TDZ.

MDA and H_2O_2 *content:* Florets and leaves exhibited low content of MDA and H_2O_2 initially but both continued to increase till 12 days in vase (Table III). The contents were very high in ethrel-treated stems and maximum in treatment with 100 µM ethrel after 12 days in vase. STS treatment did not much affect MDA and H_2O_2 content of the florets and leaves over the control. Both BAP and TDZ significantly reduced MDA content in the florets and leaves. It may, thus appear that BAP and TDZ helped in stabilizing membranes by reducing peroxidation of membrane fatty acids.

Activity of antioxidant enzymes: SOD activity of the florets showed slight increase till 6 days of storage but showed subsequent decline after that (Table 4). Treatment with ethrel was found to decrease enzyme activity where as STS had no significant effect. Treatment with BAP and TDZ increased SOD activity of florets with maximum activity (127.8 U min⁻¹ g⁻¹ FW) was observed in 10 μ M TDZ treated stems. Similar trends were observed in leaves. The activity showed increase with progress of storage duration. Ethrel suppressed SOD activity where as BAP and TDZ enhanced it. POD activity of florets showed increase in storage duration (Table 4). As a consequence, the activity reached the maximum level after 12 days of storage. Treatment with ethrel inhibited POD activity. As a consequence, high ethrel concentration (100 μ M) led to maximum decline in activity as compared to control. BAP and TDZ significantly enhanced POD activity over the control. As a consequence, the maximum activity was observed in 10 μ M TDZ treatment. The leaves exhibited lesser POD activity than the florets. The activity was minimum in leaves of ethrel-treated stems but was higher in stems treated with BAP as well as TDZ. The maximum activity was observed in stems treated with 10 μ M TDZ. Catalase activity in ethrel-treated stems where as

BAP and TDZ enhanced the same. Leaves showed continuous increase in CAT activity during storage. The activity was high in BAP and TDZ treated stems. The maximum activity was observed in leaves from stems treated with TDZ (10 μ M).

It has been suggested that metabolic processes related to senescence continue during refrigerated storage, though at a slow pace (Bhattacharjee and De 2006; Singh et al. 2012). Present studies also illustrate that with the progress of storage duration, the cut stems exhibited loss of floret longevity and early yellowing of leaves. The increase in SOD activity, which neutralizes superoxide radicals, during storage is indicative of production of ROS in florets and leaves during storage. Similarly, high activity of peroxidase and catalase during storage indicates that these enzymes play a role in removal of H_2O_2 produced in florets and leaves during storage. It has also been suggested earlier that damage induced by low temperatures could be due to generation of ROS (Campos et al. 2003; Wongshree et al. 2009; Yang et al. 2011). This may also account for increase in antioxidant enzyme activities during storage of stems.

IV. References

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Table I: Effect of ethrel, S	Table I: Effect of ethrel, STS, 6-BAP and Thidiazuron (TDZ) on days to floret senescence, yellowing of leaves and water absorption per stem in chrysanthemum var. Thai Ching Queen														
Treatments	Daystoinitiationoffloretsenescence	Daystocompletefloretsenescence	Daystoinitiationofleafyellowing	Days to 50% leaf yellowing	Total water absorbed per stem (ml)										
Control	7.33	16.67	8.67	14.67	36.7										
Ethrel, 50 µM	6.67	14.33	4.00	11.33	31.7										
Ethrel, 100 μM	5.56	10.78	3.33	10.00	30.6										
STS, 0.1 mM	7.44	16.33	7.67	14.67	33.8										
STS, 0.2 mM	8.45	16.78	8.56	15.45	35.3										
BAP, 50 μM	11.00	17.00	10.89	17.67	41.5										
BAP, 100 μM	11.56	19.67	11.67	19.00	44.7										
TDZ, 5 μM	14.89	22.44	14.33	22.78	49.7										
TDZ, 10 μΜ	15.22	23.56	14.78	23.56	51.5										
Mean	9.79	17.51	9.32	16.57	39.5										
CD at p=0.05	0.71	0.96	0.74	0.84	1.49										

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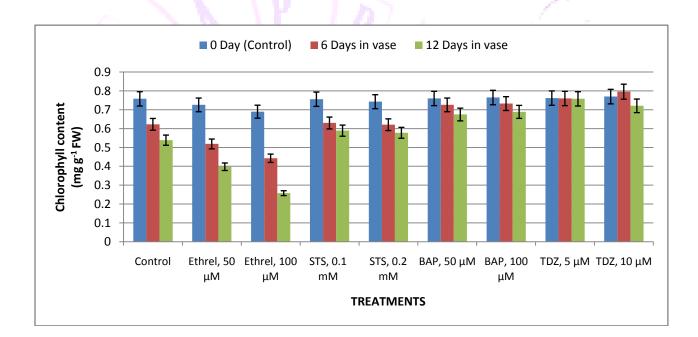


Figure I: Effect of ethrel, silver thiosulphate (STS), 6-benzyl aminopurine (BAP) and thidiazuron (TDZ) on chlorophyll content (mg g⁻¹ FW) in leaves of chrysanthemum variety Thai Ching Queen after varying durations in vase (Vertical bars represent SE of mean)

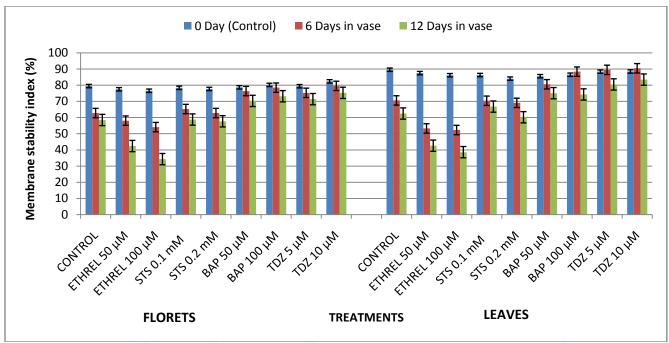


Figure II: Effect of ethrel, STS, 6-BAP and TDZ on membrane stability index (%) in florets and leaves of chrysanthemum variety Thai Ching Queen after varying durations in vase (Vertical bars represent SE of mean)



florets and lea	ves of ci	it stems	of curys	antinemu	in variety	Thai Ching	Queen alt	ler varyn	ig dura	uons m	ase						
	TOTA	L SOLU	BLE SU	GAR cor	ntent				REDU	UCING S	UGAR	content					
Treatments	FLOR	ETS			LEAVES	5			FLOR	ETS			LEAVES				
	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	
Control	522.1	355.6	264.9	380.9	148.7	116.8	81.9	115.8	48.6	24.5	18.3	30.5	6.7	4.9	3.3	5.0	
Ethrel, 50 µM	508.4	331.8	207.3	349.2	150.1	108.4	79.4	112.7	40.2	19.1	10.5	23.3	6.5	3.8	2.3	4.2	
Ethrel, 100 µM	505.5	334.8	211.3	350.5	148.9	98.2	70.2	105.8	39.6	17.5	8.9	22.0	5.9	2.8	1.2	3.3	
STS, 0.1 mM	507.7	347.6	248.5	367.9	152.3	106.6	80.8	113.2	40.9	26.7	17.6	28.4	6.6	4.2	3.4	4.7	
STS, 0.2 mM	501.3	354.3	310.2	388.6	147.8	119.8	81.7	116.4	44.4	29.5	18.7	30.9	7.0	6.6	4.6	6.1	
BAP, 50 μM	494.5	362.4	254.5	370.5	147.0	123.8	85.3	118.7	41.9	25.9	25.1	31.0	6.6	5.8	4.5	5.6	
BAP, 100 μM	493.9	371.8	274.9	380.2	151.7	133.4	90.8	125.3	47.5	31.6	28.9	36.0	8.5	6.4	6.0	7.0	
TDZ, 5 μM	502.3	370.9	263.8	379.0	153.9	125.2	91.5	123.5	44.5	42.8	39.3	42.2	7.0	7.3	7.6	7.3	
TDZ, 10 μM	507.7	364.3	283.9	385.0	151.6	135.9	107.7	131.7	45.7	47.5	40.4	44.5	8.5	10.0	10.4	9.6	
CD at p=0.05		. ,	=34.21; AXB=59			ts (A)=11.50 ; AXB=NS); Days	in vase		nents (A) B)=1.92;		•	Treatments (A)=0.53; Days in vase (B)=0.36; AXB=0.87				

Table II: Effect of ethrel, silver thiosulphate, 6-benzyl aminopurine and thidiazuron on total soluble and reducing sugar (µg mg⁻¹ DW) content in florets and leaves of cut stems of chrysanthemum variety Thai Ching Queen after varying durations in vase

Table III: Effe	ct of eth	rel, silve	er thiosu	lphate, 6	-benzyl a	aminopu	rine and	l thidiaz	aron on	H_2O_2 (m	ıM g⁻¹ F	W) and	MDA co	ontent (n	mol g ⁻¹ l	FW) in		
florets and leav	ves of cu	t stems o	f chrysa	nthemum	variety	Thai Ch	ing Que	en after v	varying d	lurations	in vase							
	H_2O_2C	ONTEN	Т					MDA CONTENT (lipid peroxidation)										
Treatments	FLORE	ETS			LEAV	ES			FLORI	ETS			LEAVES					
	0	6	12	Μ	0 6		12	Μ	0 6		12 M		0	6	12	Μ		
Control	0.543	0.673	0.865	0.693	0.507	0.754	0.853	0.705	1.643	1.993	2.353	1.996	1.130	1.357	1.933	1.473		
Ethrel, 50 µM	0.682	0.752	1.345	0.926	0.590	0.939	1.049	0.859	1.753	2.232	3.333	2.439	1.257	1.740	2.040	1.679		
Ethrel, 100 µM	0.652	1.259	1.650	1.187	0.663	1.049	1.145	0.952	1.750	2.543	3.933	2.742	1.250	2.300	2.413	1.988		
STS, 0.1 mM	0.511	0.561	0.843	0.638	0.554	0.675	0.834	0.688	1.752	2.271	2.343	2.122	1.227	1.541	2.067	1.612		
STS, 0.2 mM	0.438	0.546	0.789	0.591	0.559	0.645	0.754	0.653	1.630	2.030	2.380	2.013	1.223	1.430	1.947	1.533		
BAP, 50 μM	0.381	0.447	0.427	0.418	0.419	0.491	0.663	0.524	1.463	1.857	2.147	1.822	1.187	1.360	1.597	1.381		
BAP, 100 μM	0.334	0.433	0.574	0.447	0.389	0.473	0.642	0.501	1.400	1.637	1.837	1.624	0.977	1.150	1.213	1.113		
TDZ, 5 µM	0.353	0.452	0.558	0.454	0.403	0.539	0.724	0.555	1.033	1.315	1.453	1.267	1.160	1.2057	1.373	1.246		
TDZ, 10 μM	0.324	0.486	0.543	0.451	0.410	0.474	0.633	0.506	1.093	1.309	1.467	1.289	1.047	1.113	1.260	1.140		
CD at p=0.05		ents (A)=)=0.04; A	=0.07; XB=0.13	Days in 3		ents (A)=)=0.04; A	-	Days in		ents (A)=)=0.07; A		Days in 1	Treatments (A)=0.13; Days in vase (B)=0.08; AXB=0.23					

Table IV: Effect of ethrel, silver thiosulphate, 6-benzyl aminopurine and thidiazuron on enzyme activities in florets and leaves of chrysanthemum variety Thai Ching Queen after varying durations in vase

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eni joune	Superoxide dismutase (SOD) activity Peroxidase (POD) activity (change in Catalase (CAT) activity (mM H ₂ O)																								
	Supe	eroxic	le di	smut	ase (SOD)) acti	ivity	Per	oxidas	e (P	OD)	activi	ty (d	change	e in	Cata	lase	(CA)	F) 8	octivity	/ (m	\mathbf{M}	H_2O_2	
Treatm $(\text{U min}^{-1}\text{ g}^{-1}\text{ FW})$									abse	absorbance min ⁻¹ g ⁻¹ FW)								hydrolysed g.1 FW)							
ents	FLORETS LEAVES								FLORETS				LEAVES				FLORETS				LEA				
	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	0	6	12	Μ	
Control	98.	83	72	84	54	48	42	48	94	120	71.	95.	123	95.	77.	90.	0.5	0.4	0.4	0.4	0.9	0.8	0.7	0.8	
Control	5	.6	.6	.9	.4	.9	.4	.6	.7	.4	9	7	.8	5	8	0	46	76	35	85	81	73	53	69	
Ethrel,	93.	70	43	69	53	45	36	45	90	113	65.	89.	118	96.	78.	97.	0.5	0.4	0.3	0.4	0.9	0.8	0.8	0.8	
50µM	2	.6	.8	.2	.9	.3	.6	.3	.5	.6	5	9	.5	5	1	7	26	66	92	61	42	64	20	75	
Ethrel,	90.	68	36	65	51	38	32	41	88	110	60.	86.	112	97.	66.	105	0.5	0.4	0.3	0.4	0.8	0.8	0.7	0.8	
100µM	5	.9	.3	.2	.8	.9	.8	.2	.5	.6	5	5	.5	8	1	.5	16	43	65	41	93	10	52	18	
STS,	96.	90	70	85	52	48	41	47	93	107	87.	96.	123	107	75.	102	0.5	0.4	0.4	0.4	0.9	0.8	0.8	0.8	
0.1mM	8	.3	.3	.8	.3	.4	.7	.5	.4	.8	3	2	.4	.8	5	.2	44	88	43	91	73	46	21	80	
STS,	97.	85	75	86	53	50	48	50	95	116	81.	97.	120	102	82.	101	0.5	0.5	0.4	0.5	0.9	0.8	0.8	0.8	
0.2 mM	6	.3	.0	.0	.7	.3	.7	.9	.0	.6	2	6	.3	.3	5	.7	56	00	52	02	87	62	30	93	

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BAP,	100	95	81	92	52	58	54	55	95	131	114	113	125	112	101	113	0.5	0.5	0.4	0.5	0.9	0.8	0.8	0.8
50µM	.5	.3	.8	.5	.5	.5	.8	.3	.8	.4	.2	.8	.5	.4	.5	.1	42	13	83	12	81	92	00	91
BAP,	103	98	85	95	56	58	54	56	92	135	120	115	130	117	110	119	0.5	0.5	0.4	0.5	0.9	0.9	0.8	0.8
100µM	.2	.0	.3	.5	.7	.2	.6	.5	.1	.6	.1	.9	.6	.6	.4	.5	41	24	90	18	00	01	56	85
TDZ,	97.	97	90	95	57	62	64	61	87	127	114	109	125	128	116	123	0.5	0.5	0.5	0.5	0.8	0.8	0.8	0.8
5μM	9	.9	.5	.4	.1	.6	.9	.5	.7	.4	.6	.9	.6	.4	.3	.4	50	40	14	34	74	42	00	38
TDZ,	99.	91	88	93	58	61	65	62	94	130	120	115	128	135	120	128	0.5	0.5	0.5	0.5	0.8	0.8	0.7	0.8
10µM	5	.7	.9	.4	.6	.9	.6	.0	.3	.4	.4	.0	.9	.7	.8	.4	57	95	24	58	92	63	45	33
	Treat	tment	S		Trea	atmen	ts		Trea	tment	s (A)=	7.37;	Treatments (A)=6.03;				Treatments				Treatments			
CD at	(A)=	5.65;	Day	's in	(A)=	=5.24	; Day	/s in	Day	S	in	vase	Days		. ,	· ·	(A)=	0.008;	Day	ys in	(A)=0.020; Days in			
p=0.05	vase (B)=3.25; vase (B)=2.92;							(B)=	(B)=4.23;				Days in vase (B)=3.57; AXB=8.90			vase		(B)=0	.005;	vase (B)=0.009;				
	AXB	8=9.64	1		AX	B=8.9	AXB=9.64 AXB=8.90								(D) = 3.37, AAD = 0.90				0		AXB=0.020			

