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Water Stress and Rootstocks Influences on Hormonal Status of Budded Grapevine

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Summary

Four grape cultivars namely 'Flame Seedless', 'Thompson Seedless', 'Sharad Seedless' and 'Tas-A-Ganesh' (mutant of 'Thompson Seedless') were given different levels of moisture stress to study their physiological responses. The marginal reduction in photosynthesis and greater reduction in transpiration rate in 'Flame Seedless' have resulted in higher water use efficiency (WUE) in this variety under moisture stress. Testing of five rootstocks Dogridge, Salt Creek, 1613 C, St. George and *Vitis chamipnii* (VC) clone for drought tolerance revealed better performance of Dogridge under moisture stress conditions in terms of increased root to shoot length ratio, root to shoot dry weight ratio, increased accumulation of abscissic acid (ABA) and reduced cytokinin concentration. Budding three cultivars 'Flame Seedless', 'Thompson Seedless' and 'Sharad Seedless' on different rootstocks viz., Dogridge, Salt Creek and VC clone and subjecting to moisture stress resulted in better performance of 'Flame Seedless' and 'Sharad Seedless' budded on Dogridge in terms of increased WUE and ABA content under 50 % moisture stress. Own rooted 'Sharad Seedless' recorded least WUE under 50 % moisture stress, but when it was budded on Dogridge and subjected to moisture stress it recorded higher WUE suggesting the role of some rootstocks in manipulating the physiological changes in few scion cultivars grafted on them.

Key words. rootstocks – moisture stress – ABA – water use efficiency – root to shoot length ratio – root to shoot dry weight ratio

Introduction

Rootstock usage in Indian viticulture is realized only recently owing to the problems associated with drought and salinity in the major grape growing regions of the country. An important aspect in rootstock usage is that, when water availability is limited, root growth is generally less inhibited than shoot growth. An important feature of the root system response is the ability of some roots to continue elongating at the water potential lower than those that inhibit shoot growth (Westgate and Boyer 1985). Synthesis of abscissic acid (ABA) during moisture stress increases several folds in root system and its subsequent translocation into leaves (Bano et al. 1993). Root tips are the sites of synthesis of ABA and its synthesis increases with increased root growth. This increased root growth may enhance production of ABA. To know the extent of ABA and cytokinin synthesis in different rootstocks and also to see their accumulation in leaves of grafted vines under moisture stress situations, this study was undertaken at different levels of soil moisture stress.

Materials and Methods

Three experiments were conducted at the experimental farm of Indian Institute of Horticultural Research, Bangalore during 2002–03 and 2003–04 under open conditions. In the first experiment rooted cuttings of grape cultivars viz., 'Flame Seedless', 'Thompson Seedless', 'Sharad Seedless' (syn. 'Kishmish Cherny') and 'Tas-A-Ganesh' were planted in 14" pots containing potting mixture of red earth, sand and farm yard manure (1.5 cubic feet, approximately 15 kg) in the proportion of 2:1:1. Six-month old plants were subjected to two levels of moisture stress 50 % stress (50 % field capacity) and 100 % stress (no water) and a control with normal watering. The period of stress was 14 days. The gas exchange parameters were measured using portable open photosynthetic system (Model LCA-3, ADC, UK) before imposition of moisture stress and also on 14th day of stress cycle.

Similarly in the second experiment, rooted cuttings of five rootstocks Dogridge, Salt Creek, 1613 C, St. George and *Vitis chamipnii* (VC) clone were transplanted to the pots. Moisture stress was imposed as in experiment 1. At the end of the stress cycle, plants were uprooted from the pots and total shoot length, total root length, total shoot dry weight and total root dry weight were measured.

In the third experiment, three rootstocks viz., Dogridge, Salt Creek and VC clone were selected to study their influence on performance of three cultivars 'Flame Seedless', 'Thompson Seedless' and 'Sharad Seedless' after budding. The nine budded combinations were 'Flame Seedless' budded on Dogridge, Salt Creek, VC clone; 'Thompson Seedless' budded on Dogridge, Salt Creek, VC

clone and 'Sharad Seedless' budded on Dogridge, Salt Creek and VC clone. Six-month-old budded plants were subjected to either 50 % stress or 100 % stress for 14 days with another batch as control. Observations were recorded only in treatments of 50 % stress and control, as plants could not survive beyond 3–4 days under 100 % stress. On 14th day, gas exchange parameters and leaf ABA content were estimated.

Fully expanded young leaves were analyzed for ABA and cytokinins [zeatin riboside (ZR) and dihydro zeatin riboside (DHZR)]. ELISA technique was used to quantify ABA (Weiler 1980) and cytokinins (Barthe and Stewart 1985) employing laboratory raised polyclonal antibodies. The quantity of hormones was expressed as ng g^{-1} fresh weight of the tissue.

The treatments were arranged in Factorial Randomised Block Design with three replications. Rootstocks/cultivars and irrigation levels were fixed as factors. The comparison of treatments was carried out using t-test ($p<0.05$).

Table 1. Influence of moisture stress on gas exchange parameters in grape cultivars on 14th day of stress cycle.

Cultivars (V)	Photosynthetic rate (μmol m^{-2} s^{-1})			Transpiration rate (mmol m^{-2} s^{-1})			Stomatal conductance (μmol m^{-2} s^{-1})		
	S 1	S 2	S 3	S 1	S 2	S 3	S 1	S 2	S 3
'Flame Seedless'	10.00	9.10	*	10.50	6.90	*	0.57	0.41	*
'Thompson Seedless'	9.63	8.00	*	10.40	7.80	*	0.52	0.40	*
'Sharad Seedless'	7.50	7.06	*	10.33	8.90	*	0.42	0.39	*
'Tas-A-Ganesh'	7.83	5.73	*	10.76	9.50	*	0.43	0.36	*
Significance of F%	V	S	V×S	V	S	V×S	V	S	V×S
(P<0.05)	1.080	1.380	NS	0.371	0.321	0.643	0.042	0.037	0.074

S: Stress levels; S 1: Control (100 % irrigation); S 2: 50 % stress (50 % irrigation); S 3: 100 % stress (0 % irrigation)
*Plants wilted and hence observations were not recorded.
NS: non significant

Table 2. Influence of soil moisture stress on ABA, zeatin riboside, dihidro zeatin riboside (ng g^{-1} fresh weight) content, root to shoot length ratio and root to shoot dry weight ratio in grape rootstocks on 14th day of stress cycle.

Rootstocks (R)	ABA			Zeatin riboside			Dihydro zeatin riboside		
	S 1	S 2	S 3	S 1	S 2	S 3	S 1	S 2	S 3
Dogridge	66.37	163.83	*	43.41	32.85	*	33.56	26.60	*
1613 C	19.35	25.92	*	63.49	47.12	*	41.92	44.42	*
Salt Creek	32.46	78.05	*	52.18	34.32	*	27.85	29.70	*
St. George	12.17	31.38	*	71.16	30.92	*	52.97	30.25	*
VC clone	22.46	34.42	*	53.43	35.16	*	31.61	39.62	*
Significance of F%	R	S	R×S	R	S	R×S	R	S	R×S
(p<0.05)	5.914	3.740	8.363	3.235	2.046	4.575	3.469	2.193	4.905
	Root to shoot length ratio			Root to shoot dry weight ratio					
Dogridge	1.09	3.50	*	0.69	2.01	*			
1613 C	1.17	1.52	*	0.74	1.21	*			
Salt Creek	0.91	2.19	*	0.79	1.38	*			
St. George	0.85	1.37	*	0.98	1.10	*			
VC clone	0.75	1.99		1.09	1.22				
Significance of F%	R	S	R×S	R	S	R×S			
(p<0.05)	0.384	0.243	0.543	0.382	0.241	0.540			

S: Stress levels; S1: Control (100 % irrigation); S2: 50 % stress (50 % irrigation); S3: 100 % stress (0 % irrigation)^
*Plants wilted and hence observations were not recorded.^
NS: non significant

Results and Discussion

Photosynthetic rate, stomatal conductance (Gs), transpiration rate and instantaneous water use efficiency (WUE) recorded significant difference among the cultivars and stress levels (Table 1). On 14th day of stress cycle, maximum reduction in photosynthesis was recorded in 'Tas-A-Ganesh' from non stress to stress conditions. Among the cultivars, 'Flame Seedless' recorded maximum photosynthesis rate at 50 % stress. Considerable reduction in transpiration rate was recorded from non-stress to 100 % stress conditions. Transpiration rate was less in 'Flame Seedless' and 'Thompson Seedless' and it was highest in 'Tas-A-Ganesh'. Though there was considerable reduction in Gs with increased moisture stress, 'Flame Seedless' maintained highest Gs at 50 % moisture stress and was least in 'Tas-A-Ganesh'. 'Flame Seedless' recorded maximum WUE at 50 % stress while it was least in 'Tas-A-Ganesh'.

Significant differences in hormonal content of rootstocks at all the levels of stress were noticed. Increase in ABA content was observed with increased moisture stress and it was more in Dogridge followed by Salt Creek and VC clone. There was almost three-fold increase in ABA content in Dogridge from non stress to stress compared to other rootstocks. In contrast, there was drastic reduction in cytokinin content in all the rootstocks in response to moisture stress, which is in conformity with the concept of positive and negative root signals. This reduction was less pronounced in Dogridge and Salt Creek (Table 2). A flux of ABA from root to shoot increased only marginally in field grown Cabernet Sauvignon vines subjected to partial root drying (PRD), but it was shown in other studies that significant change in stomatal conductance (Gs) was associated with relatively small changes in bulk leaf ABA (Stoll et al. 2000). The amount of ABA accumulated in leaves may not be so important in determining the opening of stomatal aperture as Jia and Zhang (1997) have shown that it is the absolute concentration of ABA in the xylem that is more important determinant of stomatal function. This is evident in this study also that in spite of its high leaf ABA in Dogridge (163.8 ng g^{-1}) compared to other rootstocks its Gs was also highest (data not shown) leading to partial closure of stomata. The elevated concentration of ABA in xylem of PRD vines have contributed to their greater transpiration efficiency, allowing them to respond to changes in ambient evaporative demand more efficiently (Loveys 1991). In cassava, Alfredo et al. (2000) observed in addition to closing stomata, ABA promoted the characteristic developmental changes that can help plants to cope with water deficits, as restriction in shoot growth (Creeman et al. 1990) and stimulation of root extension (Sharp et al. 1994). The ABA content did not increase appreciably in rootstocks like 1613 C, St. George and VC clone with increased soil moisture stress. But, there was reduction in Gs. This might be due to hydraulic signals other than ABA produced by

Table 3. Influence of soil moisture stress on stomatal conductance, water use efficiency and ABA content in budded vines on 14th day of stress cycle.

Rootstock/Cultivar	Control (100 % irrigation)			50 % stress (50 % irrigation)		
	Dogridge	Salt Creek	VC Clone	Dogridge	Salt Creek	VC Clone
Stomatal Conductance (μmol m^{-2} s^{-1})						
'Flame Seedless'	0.64	0.73	0.74	0.27	0.36	0.43
'Thompson Seedless'	0.53	0.56	0.58	0.38	0.47	0.45
'Sharad Seedless'	0.64	0.67	0.64	0.32	0.42	0.37
Significance of F%	Rootstock	Scion	Interaction	Rootstock	Scion	Interaction
(p<0.05)	NS	NS	NS	0.060	0.060	NS
Water Use Efficiency (photosynthesis/transpiration) (μmol $mmol^{-1}$)						
'Flame Seedless'	0.85	0.92	0.82	1.09	1.09	0.71
'Thompson Seedless'	0.70	0.87	0.87	0.89	0.85	0.80
'Sharad Seedless'	0.76	0.87	0.73	1.09	0.96	0.66
Significance of F%	Rootstock	Scion	Interaction	Rootstock	Scion	Interaction
(p<0.05)	NS	NS	NS	0.137	NS	NS
Abscissic acid content (ng g^{-1} fresh weight)						
'Flame Seedless'	29.69	23.79	20.02	62.79	58.76	32.16
'Thompson Seedless'	20.21	17.75	15.59	39.06	28.07	24.38
'Sharad Seedless'	24.85	22.26	18.11	50.97	40.39	28.32
Significance of F%	Rootstock	Scion	Interaction	Rootstock	Scion	Interaction
(p<0.05)	0.866	0.866	1.500	3.024	3.024	5.238

NS: non significant

roots. NETTING et al. (1997) observed reduced Gs in stressed ABA deficient tomato mutant and they attributed it to other precursors of ABA biosynthetic pathway accumulated in the leaves, which could not be detected by the assay used for ABA. Similarly PARRY et al. (1988) reported increased accumulation of t-xanthoxin in water stressed flacca tomato mutant to similar extent as ABA in wild tomato plants. Cytokinin concentrations are often lower in xylem sap from drought-affected plants (SHASHIDHAR et al. 1996) and can influence stomatal opening. Spraying grapevine exposed to partial root water deficit with cytokinin compounds restored Gs to that of fully irrigated control (STOLL et al. 2000). In this study, the reduction in ZR and DHZR content must have contributed for significant reduction in Gs.

The increase in total root length was 2-folds in Dogridge and about 1.5-folds in Salt Creek and VC clone, while it was least in St. George and 1613 C. The reduction in shoot length was more significant in Dogridge and Salt Creek followed by VC clone (Table 2). This correlates with simultaneous increase in ABA and decrease in cytokinin at 50 % stress. The increased ABA in drying roots combined with the availability of water drawn from the wetter roots may have an impact on root growth as it has shown that grapevines subjected to PRD irrigation resulted in enhanced root development in the deeper layers, compared to control (DRY et al. 2000) and is known that ABA can maintain root growth under conditions of low soil moisture which results in inhibition of shoot growth (SHARP 1996). The reduction in the shoot development of stressed vines is due to reduction in cytokinin available from the roots (DRY and LOVEYS 1999).

Cultivars budded on Dogridge recorded higher ABA content than on Salt Creek and VC clone, which also had highest WUE (Table 3). Though own rooted variety 'Sharad Seedless' (data not shown) recorded least ABA under 50 % moisture stress, when it was budded on Dogridge it maintained highest ABA content which indicates the role of rootstocks in manipulating ABA content in scion leaves. The circulation of ABA in grafted plants was well studied by MICHELE et al. (2002) in wild type and ABA mutants of tomato with reciprocal grafting. Evidence that leaf derived ABA is exported to roots through phloem and re-circulates in xylem comes from the experiments with labeled ABA, showing that ABA synthesized in leaves appeared later in roots and xylem sap (LOVEYS 1984). There was increase in ABA content in all budded combinations from control to 50 % stress. The higher ABA content in 'Flame Seedless' and 'Sharad Seedless' on Dogridge rootstock might have contributed for reduced Gs, which in turn reduced transpiration rate and increased WUE (Table 3). Compared to ABA content of own rooted cultivars, the same cultivars when grafted on rootstocks did not show increased ABA content in same magnitude under moisture stress. This might be due to disruption of ABA translocation at bud union and also due to interaction of rootstock and scions.

References

ALFREDO, A.C., A. ALVES and T.L. SETTER 2000: Response of cassava to water deficit: leaf area growth and Abscissic acid. Crop Science **40**, 131–137.

BANO, A., K. DORFFLING, D. BETHIN and H. HAHN 1993: Abscissic acid and cytokinins as possible root to shoot signals in xylem sap of rice plants in drying soil. Aust. J. Plant Physiol. **20**, 109–115.

BARTHE, G.A. and I. STEWART 1985: Enzyme immunoassay (EIA) of endogenous cytokinin in citrus. J. Agri. Food Chemistry **30**, 293–297.

CREEMAN, R.A., H.S. MASON, R.J. BENSEN, J.S. BOYER and J.E. MELLER 1990: Water deficits and abscisic acid cause differential inhibition of shoot verses root growth in soya bean seedlings. Analysis of growth, sugar accumulation and gene expression. Plant Physiol. **92**, 205–214.

DRY, P.R. and B.R. LOVEYS 1999: Grape vine shoot growth and stomatal conductance are reduced when plants of the root system dried. Vitis **38**, 151–156.

DRY, P.R., B.R. LOVEYS and H. DURING 2000: Partial drying of the root zones of grape vines. 1. Transient change in shoot growth and gas exchange. Vitis **39**, 3–7.

JIA, W. and J. ZHANG 1997: Comparison of exportation and metabolism of xylem derived ABA in maize leaves at different water status and xylem sap pH. Plant Growth Regulation **21**, 43–49.

LOVEYS, B.R. 1984: Abscisic acid transport and metabolism in grape vines. New Phytol. **98**, 575–582.

LOVEYS, B.R. 1991: Water use is a knowledge of ABA physiology for crop improvement? In: Environment plant biology. Physiology and biochemistry of abscisic acid. Oxford, Bios scientific Publishers, pp. 245–259.

MICHELE, H.N., V.R. SHASHIDHAR, A.T. RICHARDS and R. MUNNS 2002: Stomatal control in tomato with ABA deficit mutants: Response of grafted plants to soil drying. J. Expt. Bot. **53**, 1503–1514.

NETTING, A.G., M.L. WINDSOR and B.V. MILBORROW 1997: Endogenous biosynthesis precursors of (+)-abscisic acid. Aust. J. Plant Physiol. **24**, 175–184.

PARRY, A.D., S. NEILL and R. HORGAN 1988: Xanthoxin levels and metabolism in the wild type and wilty mutants of tomato. Planta, **173**, 397–404.

SHARP, R.E., Y.Wu, G.S. VOETBERG, I.N. SAAB and M.E. LENOBLE 1994: Confirmation that abscissic acid accumulation is required for maize primary root elongation at low water potential. J.Expt. Bot. **45**,1743–1751.

SHARP, R.E. 1996: Regulation of plant growth response to low soil water potentials. Hort Science **31**, 36–39.

SHASHIDHAR, V.R., T.G. PRASAD and L. SUDARSHANA 1996: Hormones signals from the root to shoot of sunflower. Moderate soil drying increases delivery of abscisic acid and depresses delivery of cytokinins in xylem sap. Annals of Botany **78**, 151–155.

STOLL, M.B. Loveys, P. DRY and B. SHARP 2000: Hormonal changes induced by partial root zone drying of irrigated vines. J. Exptl. Bot **350**, 1627–1634.

WEILER, E.W. 1980: An enzyme immunoassay for cis-(+)-abscisic acid. Physiol. Plant. **54**, 510–514.

WESTGATE, M.E. and J.S. BOYER 1985: Osmotic adjustment and the inhibition of leaf, root, stem and silk growth at low water potentials in maize. Planta **164**, 540–549.

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