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Interaction effect of rootstocks on gas exchange parameters, biochemical changes and nutrientstatus in Sauvignon Blanc winegrapes

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ABSTRACT

SauvignonBlanc wine grape was characterized for their various morphological, physiological and biochemical parameters grafted on different rootstocks. Significant differences were recorded for all the parameters studied. The studies on vegetative parameters revealed that the rootstock influences the vegetative growth thereby increasing the photosynthetic activities of a vine. The highest photosynthesis rate was recorded in 140-Ru grafted vine followed by Fercal whereas the lowest in Salt Creek rootstock grafted vines.The rootstock influenced the changes in biochemical constituents in the grafted vine thereby helping the plant to store enough food material. Significant differences were recorded for total carbohydrates, proteins, total phenols and reducing sugar. The vines grafted on1103-Pshowed highest carbohydrates and starch followed by 140-Ru,while the least amount of carbohydrates were recorded in 110-R and Salt Creek grafted vines respectively.Among the different rootstock graft combinations, Fercal showed highest amount of reducing sugar, proteins and phenols, followed by 1103-P and SO4, however, the lowest amount of reducing sugar, proteins and phenols were recorded with 110-R grafted vines.The vines grafted on different rootstocks showed changes in nutrient uptake. Considering this, the physico-biochemical characterization of grafted vine may help to identify particularrootstocks combination that could influence a desired trait in commercial wine grape varieties after grafting.

Keywords: Gas exchange parameters, biochemical status, growth parameters,nutritional status, grape rootstocks.

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INTRODUCTION:

Grape (Vitisvinifera L)is a major horticultural crop grown on an area of about 119 thousand hectares with annual production of 21.1 million tons per hectare (Indian Horticulture database, 2013).At present,in establishing the vineyard, rootstocks is being used extensively as it provides a platform for manipulation of a broad range of vine characteristics which can consequently improve vineyard efficiency.Rootstocks have been selected to confer a wide range of other traits for grapevine improvements. These include resistance to nematodes as well as other soil borne pathogens, adaptability to soil pH (Bavarescoet al., 2003), salinity tolerance (Downton, 1977; Walker et al., 2002; Walker et al., 2004), drought tolerance (Mc Carthyet al., 1997), adaptability to water logging (Striegleret al.,1993) ability to mediate nutrient uptake and juice and wine composition (Mpelasokaet al., 2003)and the ability to control vine vigour and yield components (May, 1994; Reynolds and Wardle, 2001).

Rootstocks affect photosynthesis and dry matter partitioning of scion cultivars, which influence vegetative growth and yield. Rootstock effect on photosynthesis ingrafted vines was confirmed as scions specificby During (1994).Carboxylation efficiency is distinctively higher in all grafted varieties. The rates of photosynthesis and stomatal conductance are also influenced by rootstock genotype and age. In some cases, grafting increased the rate of photosynthesis more than it could be attributed to changes of stomatal conductance.

In addition, nutrient availability (Wong et al., 1985)and source/sink relations (Candolfi-Vasconcelos and Koblet, 1991)have been reported to affect water relations and gas exchange. Rootstocks were also found to modify leaf gas exchange of the scion under non-irrigated conditions, even though vine water status was not altered (Padgett et al., 2000). The rootstock Dogridge is performing better under Indian condition considering the yield and quality requirement. However, 110-R is another addition which is an alternative to Dogridge, looking into the soil and water problem in grape cultivation (Somkuwaret al., 2006). Considering the potential of rootstocks, the present research was carried out to study the influence of rootstocks on propagation success, vegetative growth, physiological parameters,biochemical status along with nutrients changes in Sauvignon Blancgrapevines.

MATERIALS AND METHODS

The study was conducted at the research farm of National Research Centre for Grapes, Pune during the year 2010- 2012. The experimental site is situated in mid-west Maharashtra at an altitude of 559 m; (18.32 °N and 73.51 °E). Pune has a tropical wet and dry climate with average temperatures ranging between 20 to 28 °C. The rootstocks selected for the study with their parentage as below.

Sr. No.	Rootstock used	Parentage/species
1.	110-R	V. berlandieri x V. rupestris
2.	SO4	V. berlandieri x V. riparia
3.	1103P	V. berlandieri x V. rupestris
4.	Fercal	V.berlandierixV.vinifera
5.	140Ru	V. berlandieri x V. rupestris
6.	Dogridge	V.champinii
7.	Salt Creek	V.champinii

Grafting onto rootstocks

The grafting of Sauvignon Blanc was done during September since the condition required for graft success was available during the period(high temperature 32-35°C and relative humidity between 80-90%). The grafting was done at a height of 30 cm above ground by wedge grafting method.

Vegetative growth and physiological parameters

The grafted plants were maintained following recommended standard cultural practices. The observations on growth parameters (days taken for bud sprouts, shoot length, inter nodal length, shoot diameter, leaf area and dry matter contents) were recorded at 120 days after grafting. Shoot length was measured with measuring tape while the shoot diameter was measured using Vernier calliper (0-300 mm, RSK™) at 4^{th} to 5^{th} inter nodal position. Leaf area was measured using portable leaf area meter (model CI- 203, USA). Newly matured leaf (fifth leaf from the apex) was selected to record the photosynthetic rate. Portable infrared gas analyser (model Li 6400,USA) was used to record the photosynthetic activities of leaf from the vine grafted on different rootstocks. On each plant, five leaves were selected to record the photosynthetic rate and three readings on each leaf were taken and the mean values were calculated by averaging. Chlorophyll a, b, and total chlorophyll were estimated using DMSO method.

Dry matter studies

Five shoots wereselected randomly from each graft combination and the fresh weight was recorded. These shoots were then kept for oven drying for 72 hours in hot air oven at 75°C or until no change in dry weight and dry matter percent was calculated.

Biochemical parameters

Representative leaf samples (fifth and sixth leaf from apex) from the different graft combination were obtained in triplicate. Immediately after sampling, the samples were washed with deionized water, air-dried and stored at -20 °C till extraction.

Analysis of biochemical parameters

Extractionwas carried out using the method described bySadashivam and Manickam, (1996). 0.5g of crushed sample was extracted using 10ml 80% aqueous methanol by overnight shaking at room temperature on a mechanical shaker. The supernatant was collected and residues were re-extracted twice at similar conditions. Finally, the supernatant obtained at each step of extraction was pulled together and used as a stock for estimation of carbohydrate, phenolics and reducing sugars. The residues were extracted again at 40°C with 52% perchloric acid and deionized water (two times, 30 minutes each) and the supernatant was used as a stock for starch estimation.

Carbohydrate and starch was estimated by Anthrone method while reducing sugar was estimated by the dinitrosalicylic acid (DNSA) method. Total phenolic content was estimated using Folin-Ciocalteu reagent by measuring the absorbance of the reaction mixture at 650 nm(Singleton and Rossi, 1965). The results obtained were expressed as catechol equivalent (mg/g) of the crushed sample. For protein estimation, 0.5 g of crushed samples homogenized in 0.1 M phosphate buffer (pH 7.0) was used. The homogenate was centrifuged at 5000 rpm for 15 minutes at 40°C and supernatant was used as a source for protein estimationas described byLowery method.

Nutrient status

The fully matured shoots under each rootstock combination were collected at 120 days after grafting and subjected to oven drying at 65°C for 48 hours. The shoots were then subjected to grinding, sieved and the fine powder was used for analysis of major nutrients. Among the nutrients, nitrogen was estimated using nitrogen auto analyser by Kjeldahl method using Gerhardt Distillation Unit (Vapodest 30) after digesting the samples on a digestion system. Phosphorous content in leaf was estimated using UV- visible spectrophotometer, Evolution, 201, Thermo Scientific, USA and potash content in leaf was estimated using digital flame photometer, JENWAY, UK. The nutrient content was expressed as % dry weight basis.

Statistical analysis

The experiment was conducted in randomized block design consisting of seven treatments as rootstocks. Each treatment consisted of 20 plants and was replicated three times totalling 60 plants under each rootstock. The data was subjected to calculations using the GLM procedure of SAS System software, version 9.3.

RESULTS AND DISCUSSION

Vegetative growth parameters

The data collected onvarious vegetative parameters of Sauvignon Blancgrafted on different rootstocks are presented in Table 1. Significant differences were recorded for shoot length, inter nodal length, shoot diameter and leaf area. Higher shoot length (112.0 cm), shoot diameter (7.10mm) and leaf area (178.22 cm^2) per vine was recorded in the vines grafted on Dogridge rootstock. However, increase in the fresh weight (9.36 gm) and dry weight (2.97 gm) of leaf was expressed by Fercal grafted vines. The vines grafted on Salt Creek rootstocks recorded lowest shoot length (98.00 cm), shoot diameter (5.75 m) and leaf area (142.79 cm^2). The ameliorative effect of the grafting on shoot length, shoot diameter and leaf area, dry matter percent could be attributed to the high efficacy of root system of the rootstocks in absorbing and transporting the water and minerals via the grafted union to the shoots of scion and to the favorable reciprocal relationship between stock and scion(Rafaat and Gendy, 2013). It may be concluded that, grafting on Dogridge rootstocks was beneficial asexpressed by early shoot growth that has proportionately increased total leaf area and dry matter content.In the present study, Sauvignon Blanc grafted on Dogridge rootstocks expressed more shoot length, thicker cane and maximum leaf area than the other rootstocks. Similar results were obtained byGrant and Matthews, (1996) who reported that grape cv. Krakhuna had the largest leaf surface area per vine when it was grafted on Chasselas x Berlandieri rootstock.

The observations recorded on fresh weight, dry weight and dry matter percent are presented in Table 1. Significant differences were recorded for all the parameters among the rootstocks grafted vine. It is evident from the results that rootstocks differed in their ability to accumulate dry matter in grafted vines. The maximum dry matter percentwas recorded when Sauvignon Blanc was grafted on Salt Creek (72.52%) followed by Dogridge (69.75%), whereas, the least dry matter percent (68.12%) was recorded with SO4. The study suggests that the dry matter percent accumulated in cane become available to buds for further growth of the new sprout. The results of the present study confirms the findings of Somkuwaret al.,(2009)who reported that vines of Thompson Seedless grafted on Dogridge and Salt Creek rootstocks accumulate maximum dry matter content in either canes or primary arms, which can thus be available for developing sprouts immediately after pruning. However, Sauvignon Blanc grafted on 1103-P,140-Ru, Fercal, 110-R andSO4 had minimum dry matter percent. Among the rootstocks, dry matter distribution was greatest in Salt Creek and least in SO4. The differences with the distribution of percent dry matter might be due to the genetics of the rootstocks altered with Sauvignon Blanc scions.

Gas exchanges parameters and Chlorophyll content

The data collected on various gas exchange parameters (rate of photosynthesis, stomatal conductance, transpiration rate and chlorophyll contents) of Sauvignon Blanc grafted on different rootstocks are presented in Table 2. Significant differences were recorded for rate of photosynthesis, stomatal conductance and transpiration rate. Among the different stock: scion combinations, highest photosynthesis was observed in 140-Ru (15.14 umol/cm^2/s)followed by the vines grafted on Fercal(14.32 umol/cm^2/s), while the lowest withSalt Creek rootstock grafted vines(12.74 umol/cm^2/s). The highest rate of stomatal conductance was recorded in 140-Ru followed by Fercal, SO4,Dogridge and 110-R rootstock grafted vines. The similar trend wasalso observed for transpiration rate.The presence of chlorophyll in leaf indicates the efficiency of leaf to prepare food through photosynthesis. Significant differences for chlorophyll a and b were recorded among the different rootstocks studied. The vines grafted on Dogridgerootstock recorded higher chlorophyll a (1.757 mg/g) and chlorophyll b (0.490 mg/g) followed by 110-R rootstock (1.695 mg/g) whereas, the lowest amount was noticed with Salt Creek grafted vines (1.155 mg/g). The chlorophyll b content was higher in Dogridge grafted vines than in the Salt Creek grafted vines (0.304 mg/g). The highest chlorophyll a: b ratio was observed in 1103-P grafted vines (4.129 mg/g)while the lowest in Dogridge grafted vines(3.584 mg/g). However, the higher total chlorophyll content was observed in Dogridge grafted vines (2.370 mg/g) while the lowest in Salt Creek grafted vines(1.630 mg/g).

The differences in gas exchange parameters may be due to the distinct efficacy of carboxylation of grafted vines. Similarly,During (1994) reported that the effect of rootstocks on gas exchange is a scion specific. They alsosuggested that grafting vines to appropriate rootstock favorsthe increase of carboxylation efficiency of scion leaves may help to improve droughtresistance, by raising water use efficiency.They also found that the rate of photosynthesis and stomatal conductance influenced by rootstock genotype and age.Candolfi-Vasconceloset al., (1997)found varied photosynthetic rate inV. vinifera'Muller Thurgau' grafted on different rootstocks. In their study, Pinot Noir wine grape variety exhibited higher CO_2 assimilation, transpiration rates, and higher water use efficiency when grafted on 101-14 Mgt than on 3309C rootstock.The results of the present investigation on chlorophyll contents support the findings ofBicaet al., (2000) who found that the effect of rootstock was significantly higher an chlorophyll content of grafted vine.Similarly, Keller et al., (2001) reported that the chlorophyll content was highest when vine grafted on K5BB and lowest when vine grafted on 330ac.

Biochemical status

Various biochemical constituents analyzed in the leaves of Sauvignon Blanc grafted on different rootstock are presented in Table 3. Among the different biochemical constituents, carbohydrate is considered to be important in terms of storage of vine. Variations in the total carbohydrate content in leaves of vines grafted on different rootstocks were observed in the present investigation. Higher amount of total carbohydrate was recorded in 1103-P rootstock grafted vines (98.65mg/g) followed by 140-Ru (85.75 mg/g) whereas, the least amount was recorded in 110-R (47.32 mg/g)and Salt Creek (47.65 mg/g) grafted vines.The increase in carbohydrate content in the leaf might be due to increase in leaf area that have been resulted in highest activity of photosynthesis rate which helps to synthesis more carbohydrates in the source tissue such as leaf. In the present study, the increase in leaf area might have contributed for better photosynthesis. This study supports the results obtained bySomkuwaret al., (2013)who reported potential of a vine to produce carbohydrate to meet the demands of fruit production and vegetative growth based on effective leaf area.

Starch is known to be the main reserve compound in grapevine storage tissues suchas leaves, shoots and roots. The starch content varied significantly among the different stock: scion combination (Table 3). Among the different rootstock grafted vines, the highest starch contents were recorded in Salt Creek (9.58 mg/g) followed by 1103-P (9.03 mg/g) whereas the least amount of starch was recorded in 110-R rootstock grafted vines (5.15 mg/g). The result on biochemical constituents in the present study indicates the capability of rootstock to influence different biochemical constituents in the scion which are required for physiological activation of the vine. The increased concentration of starch may be due to the decreased carbohydrate sink strength leading to accumulation of starch in leaves.Renataet al., (2010) reported that reduction in the number ofclusters probably, decreased the carbohydrate sinkstrength leading to accumulation of starch in theleaves of thinned vines. Similar results on leafcarbohydrate status were also observed in mangoleaves by Urban et al., (2004).

The protein contents in leaves of different graft combinations varied significantly.Fercalgrafted vines showed highest amount of reducing sugar, proteins and phenols, followed by 1103-P and SO4. However, the lowest amount of protein was recorded in110-R grafted vines.The results on biochemical composition exhibited significant difference due to grafting of Sauvignon Blanc onto different rootstocks. This might be due to the alterations in the growth pattern of vines by rootstocks as well as the differences in their uptake of nutrients and water from soil solution, as root development patterns vary with the rootstocks. Most secondary, effects of rootstocks are mediated through their influence on vine size and internal canopy shading. The results of the present investigations confirms with the results obtained by Mabrouk and Sinoquet (1998), who reported that canopy structure and sunlight exposure had positive relationship with phenolic contents. In addition, a relationship between variations in vine growth and differences in total phenolic levels has also been reported byLamb et al.,(2004) andCortellet al., (2005).

Nutrient status

The data recorded on nutrient status in the vines grafted on different rootstocks are presented in Table 4. Among the different nutrients, Nitrogen (N) is one of the major nutrients required by plants for vegetativegrowth. Whenit applied ininsufficient or excessive amounts, it can cause negative effects in plant production and productivity. Considering the effect of rootstock on leaf nutrient contents, it wasobserved that 140-Ru and Fercal rootstocks grafted vines were the most efficient innitrogen and potassium uptake but had anintermediate performance for the uptake of phosphorus,while Salt Creek ranked as the efficient rootstock in phosphorus uptake as compared to other rootstocks. The reports on mineral uptake and distribution in grapevinesconcluded that the differences in nutrient uptake and distribution could be attributed to the genotype of rootstock which gives different absorption capability or tendency for some specific minerals. These differences are due to the interspecific variation among the rootstocks in terms of nutrient absorption as reported by Grant and Mathews, (1996).The variation in phosphorus uptake have also been reported by Ruhlet al., (1998); Bavarescoet al.,(2003) and Troncosoet al.,(1999),who suggested that the different rootstock absorb unlike levels of phosphorus with concomitant effects on the growth of shoots and leaves.

Maximum K was accumulated in Fercal grafted vine(1.25%) followed by Dogridge rootstocks (1.11%) and least K were found in SO4 grafted vines (0.78%). The differences in accumulation of K in vines may be due to the effect of individual rootstock. The result also indicates that the mechanisms of accumulation of K in scion are controlledby rootstocks.Fisarakiset al., (2004) andMpelasokaet al.,(2003) reported that the variation could be caused by differences in the incorporation from the roots to shoots. Ibacache and Sierra (2009) reported that differences among the rootstocks in the morphology and density of the roots in the soil profile could also explain the variations in the K absorption capacity of the roots. The results of the present investigation for variation in the level of nutrient in Sauvignon Blanc grafted on different rootstocks might be due to the differences in the rooting pattern and the physiology of individual rootstock. The difference in level of nutrients in grafted variety has also been reported by several workers (Troncosoet al., 1999; Bavarescoet al., 2003; Garcia et al., 2001 and Robinson, 2005).The results of the present study also supports the findings ofFardossiet al., (1995) who reported the accumulation of potassium, calcium, magnesium and phosphorus in different rootstock grafted vine is variety and season specific. Keller et al., (2001) reported that the results obtained with a particular stock: scion combination in a specific environment cannot be extrapolated to other situations. Jackson (2000)reported that the interaction usually results from the mutual translocation of nutrients and growth regulators between the scion and rootstock.

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Table (1): Effect of different rootstocks on vegetative growth parameters in Sauvignon Blanc vines

Rootstock	Shoot length (cm)	Inter nodal Length (cm)	Shoot Diameter (mm)	Leaf Area (cm^2)	Fresh Wt(gm)	Dry Wt(gm)	Dry Matter (%)
110-R	94.89^d	5.10^{de}	6.05^d	157.33^c	9.57^a	3.02^a	68.47^c
SO4	104.22^c	5.40^b	6.78^{bc}	165.33^b	8.54^b	2.72^b	68.12^c
1103-P	110.33^b	5.60^a	6.10^d	160.00^c	7.43^e	2.15^d	71.11^{ab}
Fercal	96.00^d	5.45^b	6.97^{ab}	168.55^b	9.36^a	2.97^a	68.28^c
140-Ru	115.0^a	5.05^e	6.65^c	165.67^b	8.03^d	2.44^c	69.63^{bc}
Dogridge	112.0^{ab}	5.20^{cd}	7.10^a	178.22^a	8.32^c	2.50^c	69.95^{bc}
Salt Creek	98.00^d	5.25^c	5.75^e	142.79^d	6.28^f	1.73^e	72.52^a
CV %	2.145	1.539	1.646	1.730	1.498	1.705	1.503
LSD 5 %	3.982	0.145	0.19	5.004	0.219	0.076	1.864
Significance	**	**	**	**	**	**	**

Table (2): Effect of different rootstocks on gas exchange parameters and chlorophyll content in Sauvignon Blanc vines

Rootstocks	Photosynthesis (umol/cm^2/s)	Stomatal Conductance (cm·s-^{1})	Transpiration rate (mmol H_2O m^{-2}s^{-1})	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Chlorophyll a : Chlorophyll b	Total Chlorophyll (mg/g)
110-R	13.81^c	0.240^d	3.565^f	1.695^{ab}	0.420^b	4.035^{ab}	2.249^b
SO4	14.43^b	0.272^b	3.891^d	1.301^c	0.338^d	3.853^c	1.799^c
1103-P	14.04^{bc}	0.247^{dc}	3.722^e	1.654^b	0.401^c	4.129^a	2.193^b
Fercal	14.32^b	0.281^b	4.154^c	1.646^b	0.400^c	4.112^{ab}	2.185^b
140-Ru	15.14^a	0.330^a	5.074^a	1.134^d	0.282^f	4.022^b	1.591^d
Dogridge	13.3^d	0.255^c	4.081^c	1.757^a	0.490^a	3.584^d	2.370^a
Salt Creek	12.74^e	0.276^b	4.642^b	1.155^d	0.304^e	3.794^c	1.630^d
CV %	1.628	1.882	1.938	2.408	2.606	1.488	2.247
LSD 5 %	0.407	0.009	0.143	0.063	0.017	0.104	0.080
Significance	**	**	**	**	**	**	**

Table (3): Effect of different rootstocks on biochemical constituents in Sauvignon Blanc vines

Rootstocks	Reducing Sugar (mg/g)	Proteins (mg/g)	Phenols (mg/g)	Starch (mg/g)	Total Carbohydrate (mg/g)
110-R	18.56^e	14.50^e	9.11^d	5.15^f	47.32^f
SO4	32.13^d	18.20^b	9.74^b	6.12^e	54.98^e
1103-P	47.65^b	17.42^c	9.04^d	9.03^b	98.65^a
Fercal	50.19^a	19.70^a	10.76^a	7.73^d	64.98^d
140-Ru	32.13^d	16.84^d	9.15^d	8.44^c	85.75^b
Dogridge	31.41^d	14.79^e	8.21^e	7.90^d	78.65^c
Salt Creek	45.41^c	16.94^d	9.38^c	9.58^a	47.65^f
CV %	3.285	1.517	1.041	2.820	4.328
LSD 5 %	2.149	0.456	0.173	0.386	5.258
Significance	**	**	**	**	**

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Table (4): Effectof different rootstockson nutrient status in Sauvignon Blanc vines

Rootstocks	N %	P %	K %	Na (%)	Ca (%)	Mg (%)	Mn (ppm)	Cu (ppm)	Fe(ppm)	Zn (ppm)
110-R	0.69^{c}	0.11^{d}	1.013^{c}	0.294^{b}	0.68a	0.270^{c}	36.60^{d}	10.50^{c}	116.00^{c}	62.85^{de}
SO4	0.60^{e}	0.11^{d}	0.788^{d}	0.319^{a}	0.67ab	0.290^{b}	58.80^{a}	12.70^{a}	93.40^{g}	75.85^{b}
1103-P	0.76^{b}	0.09^{e}	1.00^{c}	0.263^{c}	0.65b	0.300^{b}	53.40^{b}	11.10^{b}	105.40^{e}	64.65^{cd}
Fercal	0.71^{c}	0.13^{c}	1.25^{a}	0.231^{d}	0.67ab	0.230^{d}	40.15^{c}	9.40^{d}	121.10^{b}	61.00^{ef}
140-Ru	0.83^{a}	0.13^{c}	0.988^{c}	0.219^{e}	0.60c	0.320^{a}	34.70^{d}	8.60^{e}	102.65^{f}	65.05^{c}
Dogridge	0.69^{c}	0.14^{b}	1.113^{b}	0.225^{de}	0.60c	0.290^{b}	22.85^{f}	13.00^{a}	109.55^{d}	60.75^{f}
Salt Creek	0.64^{d}	0.18^{a}	0.750^{e}	0.194^{f}	0.44d	0.220^{d}	29.95^{e}	5.70^{f}	123.70^{a}	77.90^{a}
CV %	2.166	1.971	2.080	2.389	1.946	2.618	3.921	3.288	1.091	1.569
LSD 5 %	0.027	0.004	0.036	0.010	0.021	0.012	2.755	0.593	2.140	1.866
Significance	**	**	**	**	**	**	**	**	**	**