



Influence of different grape rootstocks on rooting behavior, photosynthetic activities and biochemical constituents in different parts

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ABSTRACT

Twelve grape rootstocks were characterized for their various morphological, physiological and biochemical parameters. Significant differences were observed for most of the parameter studied. Rootstocks such as 110-R, 99-R, Dogridge and 1103-P had significant higher contents of all major biochemicals. The highest rate of photosynthesis and rate of transpiration at single leaf level was recorded for 110-R and Salt Creek rootstocks which is an important mechanism in overcoming drought tolerance. Highest total Phenolic in the rootstocks such as Dogridge, 110-R and V. longii, may help in reducing the incidence of major grape diseases in profitable table varieties, if grafted onto these rootstocks. Thus, the physio-biochemical characterization of rootstock may help to identify particular rootstocks that could influence a desired trait in commercial table or wine grape varieties after grafting. In the present investigation, the rootstocks such as 110-R, 99-R 1103-P and Dogridge recorded the highest value for total carbohydrates, phenols, proteins, reducing sugar and gas exchange parameters which may help these rootstocks in overcoming the incidence of important disease and drought tolerance and lead to a better rooting percentage.

Keywords : Gas exchange parameters; biochemical status; growth parameters; grape rootstocks.



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INTRODUCTION

Grape (*Vitis vinifera* L.) is one of the important fruit crops cultivated widely in temperate and subtropical climates. Even though their origin was in temperate regions, they perform equally well in a tropical climate in India, where they are grown as an evergreen vine without undergoing dormancy. Rootstocks are used in most grape growing countries to overcome biotic stresses like nematodes, phylloxera, root lice, etc. (Satisha *et al.* 2007). In India, however, rootstocks are gaining popularity because of their tolerance to abiotic stresses like drought and salinity. Under Indian conditions, Thompson Seedless is the most popular commercial variety and is grown in larger areas. But this variety is highly susceptible to major diseases, like downy mildew, powdery mildew and anthracnose. Hence, grafting on suitable rootstock can be the only alternative to obtain sustainable yield. Extensive grafting experimentation has revealed that rootstocks exert their influence on several aspects of scion physiology (Rosa *et al.* 2003). Because of affinity of rootstocks for the scions, the choice of appropriate rootstock is of great importance for the quality of grafted grapevines. As the physiology and biochemistry of rootstocks vary under similar sets of management practices, the physiology and biochemical composition of the mother vines play an important role in the propagation, growth and development of vine, quality of the grapes etc. As grape rootstocks belong to different *Vitis* species, each rootstock has its own inherent capacity to synthesis biochemical constituents, which influence scion physiology, either directly or indirectly, after grafting (Satisha and Prakash 2006). Some may influence drought tolerance through the accumulation of osmolytes, increasing water-use efficiency, while some may influence disease and pest resistance through the accumulation of polyphenols, phytoalexins, etc. (Satish *et al.* 2007). Considering these, the present investigation was carried out to categorize the rootstocks on the basis of various gas exchange parameters, its rooting behavior and status of biochemical in different parts.

MATERIALS AND METHODS

The study was carried out in the experimental nursery of National Research Center for Grapes, Pune during the year 2011 and 2012. Pune is situated in mid-west of Maharashtra at an altitude of 559m above mean sea level; (18.32 °N and 73.5 1 °E). Pune has a tropical wet and dry climate with average temperature ranging between 20 to 28 °C. Well matured lignified canes from different rootstocks (Table 1) were harvested from 8-year-old mother vine and the cuttings with four buds were prepared and bundled. These cuttings were then kept in tap running water for overnight so as to leach out the rooting inhibitors. After removal from water, a slanting cut at the basal end was given to expose more area for rooting. The cuttings of different rootstocks were treated with 1000 ppm IBA for better rooting and planted in poly bags of 7" x 4" size and irrigated immediately.

Vegetative parameters

Days taken for bud sprout were recorded from the date of planting to sprouting. The first sprouted bud with fully expanded leaf was taken as an indicator to calculate the days taken for sprouting. The vegetative parameters such as shoot length were measured by measuring tape; shoot diameter and internodal length by digital Vernier caliper at 4th to 5th internodal position at 90 days after planting. For physiological and biochemical studies, the fully developed and recently matured leaf were selected (usually the 5th/6th leaf from the apex).

Biochemical parameters

The samples from leaf, cane and roots were collected randomly from five mother vines of each rootstock. The samples were washed thoroughly with distilled water, air-dried and stored at -20°C. Each sample was subsequently lyophilized using a freeze drier (Benchtop 4 K VIRTIS) at -78 °C. The lyophilized samples were blended thoroughly and sieved through a sieve with 40 mesh size and stored at -20°C until further processing.

Extraction of samples

One gram each of the different samples lyophilized in three replications was extracted by overnight shaking at room temperature on a mechanical shaker in the dark. The solvent used was 80% aqueous methanol, as this has been reported to be a better solvent for biochemical extraction (Bonilla *et al.* 2003). The mixture was centrifuged at 12000 rpm for 15 minutes at 4°C. The residues were re-extracted (three times for three hours each) under similar conditions. The complete leaf extraction was ensured by a qualitative Folin-Ciocalteu negative test on Whatman filter paper No. 1. The filtrates were pooled and concentrated to one-third of their volume using Turbovap concentrator under a gentle stream of nitrogen. The sample extracts were treated with chloroform to remove chlorophyll, and residual aqueous extracts were washed away with ethyl acetate (Park and Cha 2003). The extracts were filtered through 0.45 mm filters and stored at 0°C until further analysis (Ju and Howard 2003).

Estimation of Biochemicals

The total carbohydrates content on dry weight basis in different parts (leaf, shoot and roots) was estimated as per Sadashivam and Manickam (1992) and was expressed as mg/gm. Protein estimation was carried out as per Lowry's method and the estimated protein was expressed in mg g⁻¹ sample. The reducing sugar was estimated by following Dinitrosalicylic acid method and was expressed in terms of mg g⁻¹. The total polyphenol content of the extract was determined using the Folin-Ciocalteu method (Singleton and Rossi 1965), using Gallic acid as the standard. The concentration of the total phenolics was expressed as the gram equivalent of the sample.



Leaf chlorophyll Contents

Mature leaves of each rootstock were collected from all sides of the mother vine, washed initially with tap water followed by watering with distilled water. The leaf chlorophyll contents (a and b) were determined as per the method suggested by (Moran and Porath 1980) and was expressed in mg g^{-1} of sample.

Gas exchange parameters

Gas exchange parameters such as photosynthetic rate and transpiration rate in leaf were measured using infrared gas analyzer (IRGA) in the leaves before they were sampled for the estimation of the various biochemical constituents. The observations were recorded in full sunlight between 10:00 and 11.30 am. The area of the chamber used for holding the leaves was 6.25 cm^2 . The photosynthetic rate was expressed as $\text{mol CO}_2/\text{m}^2/\text{sec}$, while the transpiration rate was expressed as $\text{mmol H}_2\text{O}/\text{m}^2/\text{sec}$. Water-use efficiency at the level of a single leaf was derived using the formula $\text{WUE} = \text{photosynthetic rate} / \text{transpiration rate}$.

Statistical Analysis

Analysis of variance was performed for each variable using the SAS statistical package 9.3 (SAS Institute, Cary, NC). Least significant differences among treatments were calculated.

RESULTS AND DISCUSSION

Vegetative parameters

The data collected on various vegetative parameters of different rootstocks are presented in Table 2. Significant differences were recorded for days to bud sprout, percent rooting and other vegetative parameters (shoot length, diameter and internodal length). The rootstock V. Champinii sprouted earlier (7.0 days) followed by 99-R (8.0 days) and St. George (8.2 days) compared to the late sprouting in Freedom rootstocks (13.0 days). Higher shoot length (45 cm), shoot diameter (5.30 mm) and percent rooting (81.40) were also recorded in St. Gorge rootstock. The early bud sprout and increased vigour through shoot length in this rootstock may be attributed to the increased activity of polyphenol oxidase in their buds. Satisha *et al.* (2012) reported positive correlation between bud burst and PPO activity. The period for bud burst in these rootstocks was in accordance with the earlier report of several workers who established the influence of rootstock on bud burst. Prakash and Reddy (1990) reported significant differences on bud sprout throughout different rootstock. The biochemical changes in different parts of vine during bud break have been studied by several workers (Kenis 1979; Marqu *et al.* 1999; Sivaci 2006). The change in enzyme activity seems to be an indicator of the end of dormancy and the start of growth, as described by few researchers (Baasuk *et al.* 1981; Citadin *et al.* 2011).

The vegetative growth parameter such as average shoot length, shoot diameter and internodal length showed significant differences among the different rootstocks. The vegetative growth in terms of shoot lengths was recorded on Dogridge and St. Gorge rootstocks (45.0cm) whereas the rootstock 110-R had reduced vigour (30.0 cm). However, moderate vigour was recorded in salt Creek, SO4, V. Champinii and 1103-P rootstocks. Compared to other rootstocks, the increased vigour of Dogridge is attributed to more vigorous growth, which is evident from increased root length; shoot diameter, scion girth after grafting in the field as reported earlier by Shobhana *et al.* (2000).

The data on percent rooting recorded among the different rootstocks are presented in Table 2. Among the different rootstocks, the highest rooting percentage was recorded in the rootstock V. longii and Salt Creek (82.00 % each) followed by St. George (81.40 %) while the rootstock 110-R exhibited least rooting percentage (77.00 %). The lower percent rooting for 110-R rootstocks might be due to the availability of a higher percentage of rooting inhibitors. Sturve (1981) reported that a higher C: N ratio in the tissues of cuttings promotes the rooting. The results obtained in the present investigation can be attributed that the reserve food materials might play an important role in rootlets of different rootstocks. The higher percentage of rooting in Freedom rootstock was found to be associated with the increased amount of carbohydrates. The supply of carbohydrate while rooting might have helped to produce more rootlets and thus increased root length in the Freedom rootstock. The results obtained in this study are in accordance with the result obtained by Sidhu and Bose (1982), who reported that, the level of soluble sugar and C: N ratio positively correlated with rooting in guava.

Biochemical constituents in leaf

The various biochemical constituents analyzed in the leaves, shoots and roots of different grape rootstocks are presented in Table 3. Among the different constituents, carbohydrate is considered to be an important in terms of storage of vine. In leaves, total carbohydrates were highest in Dogridge rootstock (138.88 mg/g), followed by Salt Creek (124.96 mg/g) and SO4 (117.54 mg/g). The lowest carbohydrate content was recorded in 110-R (63.61 mg/g), St. George (62.46 mg/g) and 1613-C (69.24 mg/g) respectively. The increase in carbohydrate content in leaf might be due to increase in canopy with 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 by increase in number of shoots might have contributed for better photosynthesis. This study supports the results obtained by Somkuwar *et al.* (2013) who reported that potential of a vine to produce carbohydrate to meet the demands of fruit production and vegetative growth based on leaf area.

Starch is known to be the main reserve compound in grapevine storage tissues such as leaves, shoots and roots. The starch contents in the leaves varied significantly among the different rootstocks with highest in SO4 (17.19 mg/g) and was followed by Freedom (17.01 mg/g), Dogridge (15.22 mg/g) and Salt Creek (14.69 mg/g) respectively. The least amount of



starch in the leaf was recorded with *V. champinii* (3.79 mg/g) followed by 110-R (8.17 mg/g). In the present findings, the increased concentration of starch may be due to the decreased carbohydrate sink strength leading to accumulation of starch in leaves. The present study confirms the findings of Renata *et al.* (2010), who reported that reduction in the number of clusters probably decreases the carbohydrate sink strength leading to accumulation of starch in the leaves of thinned vines. Similar results on leaf carbohydrate status were also observed by Urban *et al.* (2004) in mango leaves.

Among the different rootstock studied, highest protein contents was recorded in the leaf of 110-R (114.33 mg/g) followed by 99-R (94.17 mg/g) and Freedom (88.0 mg/g), whereas, the lowest (44.67 mg/g) amount of protein was recorded in *V. champinii*. The data showed major differences in the protein contents among the different grape rootstocks indicating the existence of wide range of variation. The changes in the protein content among the different rootstock might be due to the response of individual rootstock. Factors including cultivar, rootstock/scion combination, vine nutrient management, vineyard site and growing season affect the proteins and amino acid concentration within the grapes (Gump *et al.* 2002; Bell Henschke 2005).

The phenols being considered major biochemical constituents in grapevine. The phenolic contents in leaves of different rootstocks ranged from 72.70 mg/g to 196.0 mg/g. The higher phenol content was recorded in the leaf of 110-R rootstock (196.0 mg/g) and 140-Ru (192.52 mg/g) rootstock. However, the rootstocks *V. champinii* exhibited lowest quantity of phenols in leaf (72.70 mg/g) sample. The present findings are in accordance with findings of Satisha *et al.* (2007) who concluded that the rootstock in the group of *V. berlandieri* x *V. rupestris*, such as 110-R recorded the highest values of total phenols and protein. Increased phenol content may help to reduce the diseases incidence in grapevine. Similarly, Somkuwar *et al.* (2014) while working on Thompson Seedless grafted on Dogridge and 110-R rootstock, reported less disease incidence of anthracnose with higher phenol contents in leaf of grapevine.

The reducing sugar ranged from 30.44 mg/g in *V. chapinii* to 90.78 mg/g in SO4 leaf while *V. longii*, 99-R, 1613-C, Freedom and 110-R were ranged inbetween the range Table 3. The variations for the reducing sugars might be related to the changes in the photosynthetic activities of vine. The results of this study also confirms the findings of Somkuwar *et al.* (2013) who reported that positive correlation between photosynthetic activity and reducing sugar.

Biochemical constituents in Shoots

In grapevine, shoots/ cane are considered as one of the major plant part storing carbohydrates as food material which can supply to the developing bunches after fruit pruning. The carbohydrate contents in the shoot varied significantly among the different rootstock (Table 3). The carbohydrate content ranged from minimum of 300.20 mg/g in 1103-P to highest in 110-R rootstock (736.93 mg/g). Highest amount of carbohydrate content in 110-R may help the vine better storage of food material leading to higher yield per vine. The increased carbohydrate contents may be due to the increased canopy for active photosynthesis stored in new cane. Similar results were obtained by Omar *et al.* (2000) in Thompson Seedless and Crimson Seedless. El-Baz *et al.* (2002) also studied influence of pruning severity on bud behavior, yield, berry quality and some biochemical contents of canes of Crimson Seedless grapes.

Starch contents in shoot were significantly varied among the different rootstocks with maximum in Dogridge (19.24 mg/g) followed by *V. champinii* (122.54 mg/g) and Salt Creek (103.35 mg/g). However, the rootstock 110-R exhibited lowest amount of starch (32.37 mg/g) in shoot as compared to all rootstocks. The increased starch concentration coincided with the maximum expansion of leaf area. Uys and Orffer (1983) observed that the increased concentration of starch in Jacquez stems was more pronounced than in Salt Creek.

Highest amount of protein was recorded in shoots of Dogridge rootstock (442.47 mg/g) followed by SO4 (382.80 mg/g) and 99-R (366.80 mg/g). However, the lowest protein content in 1103-P (262.13 mg/g) was recorded in the present investigation. The data showed major differences in the protein accumulation pattern among the different grape rootstocks indicating the existence of wide range of variation. The quantity of protein was found to be more specific in different rootstocks. The results clearly showed that individual rootstocks have the capacity to synthesize their food differently. As the phenology and biochemistry of rootstock vary under similar set of condition, the biochemical as well as physiological composition of mother vine might be playing role in propagation, growth and development of a vine, water use efficiency, pest and disease tolerance and ultimately the quality of grapes (Staudt 1997). Satisha and Prakash (2006) in their studies also reported inherent capacity of each rootstock to synthesize biochemical constituents, which influence scion physiology either directly or indirectly after grafting.

The status of phenol content in the shoots of different rootstock varied significantly with higher amount in Dogridge (243.37 mg/g), followed by Salt Creek (131.45 mg/g); while the lowest quantity was recorded in 140-Ru (28.50 mg/g). In the present investigation, the rootstock 1103-P and *V. longii* had higher phenol with maximum rooting percent. Several researchers have reported that phenolics are negatively related to seed germination and in vitro proliferation (White 1994; Prasad 1989). Phenolic compounds sometimes have an inhibitory or stimulating effect on plant growth, which varies from species to species (Ozyigit *et al.* 2007). The present study indicated that the rooting behavior and phenolic contents varies from rootstock to rootstock and it is largely dependent on the genetic make-up of the rootstock. The variation in phenolic content of shoot might be due to the variation in genetic make-up of mother vine. The findings of present investigations are in accordance with our earlier findings of variation of phenolic content from one rootstock to another (Somkuwar *et al.* 2012). Reducing sugar varied significantly among the different rootstocks with higher amount in 99-R rootstock (55.80 mg/g) than the least in Salt Creek (23.91 mg/g). The changes in the reducing sugar content might be due to the changes in the photosynthetic activities of vine (Somkuwar *et al.* 2013).



Biochemical constituent in Roots

The roots of grapevine play an important role in absorbing the nutrients and water from the soil to supply the aerial parts of canopy. It also acts as a storage organ which supplies the food material to the vine during the growth period. The different biochemical constitute studied in roots of different rootstock is presented in Table 3. The variation in Carbohydrate content was recorded with maximum in *V. longii* (860.27 mg/g) followed by 99-R (752.23 mg/g), whereas the lowest amount of carbohydrate was recorded in Salt Creek (447.77 mg/g). The starch content in roots was significantly varied from 279.24 mg/g in 99-R to 66.29 mg/g in Freedom rootstock. Protein content in roots ranged from 974.17 mg/g in 140-Ru to 452.50 mg/g in Freedom rootstock. It is assumed that the increase in starch content in root may be due to the leaves which were photosynthetically active and continued to supply carbohydrates to storage tissue. This confirms the findings of Scholefield *et al.* (1978). The low starch content in roots of grapevine may reflect the high contents of carbohydrate demand of vegetative growth. Hunter *et al.* (1995) found that starch built-up from berry set to post harvest stage coincides with the pattern in leaves. This indicates that carbohydrates availability increases during the vegetative period and carbon partitioning between leaves and shoots is interrelated. Close relationships between above-ground and subterranean growth of grapevines are known to exist (Hunter *et al.* 1995).

The trend in variation in phenol content in roots of different grapes rootstocks was also observed in the present study. The higher amount of phenol was recorded in the roots of 1103-P followed by *V. longii* (704.35 mg/g) whereas, the lowest phenol contents (375.65 mg/g) were recorded in Freedom rootstock. Reducing sugar in the roots of different rootstocks varied significantly with highest amount in 140-Ru (79.44 mg/g), followed by 99-R (75.00 mg/g) and the lowest (40.0 mg/g) in Freedom.

Among the different plant parts studied for various biochemical parameters (carbohydrates, starch, proteins and phenol) was higher in Dogridge rootstock followed by Salt Creek, 110-R and *V. longii*. Several workers believe that the characterization of germplasm based on biochemical composition is useful in identifying synonyms and helps in identifying the potential accession for a given trait. Aseusio *et al.* (2002) characterized white wine grape cultivars grown in Extremadura region of Spain at various phenological stages and characterized the amino acid composition and able to differentiate the varieties. They could also identify the commoners among and the similarities between the cultivars and found that berry protein could be used as a biochemical marker to identify genetic variation in Muscadine cultivars.

In the present investigation, we could observe the trend where the accession belong to same group such as 110-R, 1103-P and 99-R had a similar biochemical composition than those belongs to other species. The rootstocks can thus be categorized based on the phenol accumulators, high carbohydrate and reducing sugar accumulators and thus it may becomes easy to standardize the propagation practice for a specific rootstock to achieve better rooting and growth success.

Though, in the present study, influence of these rootstocks on physiology and biochemistry of scion after planting, the inherent capacity of rootstock will still have the positive influence after grafting. Several reports are available on the status of biochemical composition of scion in various species, such as on apple (Brown *et al.* 1985), sayabean (Caver *et al.* 1987). In Hevea, it has been found that rootstocks have profound influence on the biochemical composition of the leaves, especially in terms of reducing sugar, phenols and protein (Shobhana 1998).

Gas exchange parameters and chlorophyll contents

Gas exchange parameters play an important role in determining the ability of plant to photosynthesis and utilize water efficiently at the level of a single leaf. This is considered as one of the important drought tolerance mechanisms in most of the crop species. The gas exchange parameters studied in different rootstocks are presented in Table 4. The higher photosynthetic rate was recorded in Salt Creek (16.07 $\mu\text{mol}/\text{mg}/\text{s}$) followed by 99-R (14.87 $\mu\text{mol}/\text{mg}/\text{s}$), St. George (14.84 $\mu\text{mol}/\text{mg}/\text{s}$) and 110-R (14.80 $\mu\text{mol}/\text{mg}/\text{s}$). However, the rootstock 140-Ru recorded the lowest photosynthetic rate (7.38 $\mu\text{mol}/\text{mg}/\text{s}$) as compared to other rootstocks studied. The increase in photosynthetic activities may help the vine to improve storage thereafter increasing the yield. The findings of the present investigation may correlates with the earlier findings of increase in yield of Thompson Seedless grafted on 110-R rootstock followed by and Dogridge (Anonymous, 2006). The significant differences were also recorded for stomata conductance and transpiration rate. Stomatal conductance was higher in Salt Creek (0.421 mm/s) followed by *V. Champinii* (0.403 mm/s), while the lowest stomatal conductance (0.076 mm/s) was observed in 140-Ru rootstock. The maximum transpiration rate (3.700 mm $\text{H}_2\text{O m}^{-2}\text{s}^{-1}$) was recorded in the Dogridge rootstock, followed by Salt Creek (3.59 mm $\text{H}_2\text{O m}^{-2}\text{s}^{-1}$), whereas, the least rate of transpiration (1.300 mm $\text{H}_2\text{O m}^{-2}\text{s}^{-1}$) was recorded in 140-Ru rootstock. The reduction in stomata conductance might limit water vapor loss via transpiration which may help to avoid drought. The present study also confirms the results obtained by Kirkham (1990) and Passioura (1994) who worked on plant response to water deficits and the yield of crop in relation to drought response.

The data recorded on chlorophyll a, chlorophyll b, chlorophyll ab, and total chlorophyll are presented in Table 4. Among the different rootstocks, 110-R showed maximum (2.115 mg/g) chlorophyll a, (0.612 mg/g) chlorophyll b and also total chlorophyll (2.820 mg/g), followed by Salt Creek rootstock. The least amount of chlorophyll a (1.101 mg/g) and total chlorophyll (1.653 mg/g) was recorded in *V. Longii*, while the rootstock SO4 showed minimum chlorophyll b (0.446 mg/g) and chlorophyll ab (2.910 mg/g). The increased chlorophyll contents in 110-R and Salt Creek may be due to the increase in percent root and also shoot diameter helps the plant to grow at a faster rate. The presence of starch, proteins and carbohydrates in the rooted plants acts as an energy source for further growth. In 110-R and Salt Creek rootstock, higher amount of carbohydrates, protein, reducing sugar were recorded when compared to other rootstocks. These rootstocks might have availability of biochemical constitute at higher rates, which helped the plant to grow via the production of more



chlorophyll required for photosynthesis. The present findings confirms the study of Zachariakis et al. (2001) who reported that total chlorophyll content increased the total carbohydrate concentration in grapevine shoots.

In the present study the trends for biochemical accumulation indicate that the rootstocks 140-Ru, 110-R and Freedom accumulated maximum phenol in leaves, 1103-P, V. longii and 140-Ru accumulated more phenol in roots whereas the rootstocks Dogridge and Salt Creek accumulated more phenols in their shoots. The minimum phenol accumulation in leaf was observed in V. Champinii, Dogridge and 1103-P whereas the lower phenol accumulation in shoot was recorded in 140-Ru, St George and 1613-C rootstock and the rootstock Freedom, 1613-C, St George recorded less phenols in roots. Accumulation of higher phenol in these rootstocks might help in developing the resistance/tolerance to disease and pest.

The rootstocks such as Dogridge, Salt creek and SO4 accumulated maximum carbohydrates in leaves while the rootstocks 1613-C, 110-R, 140-Ru showed minimum carbohydrates in their leaves. However, the rootstocks 110-R, Freedom and Salt Creek accumulated higher carbohydrates in shoots while the rootstocks 1103-P, V. Longi and V. Champanii accumulated least carbohydrates in the shoots. On the other hand, the roots of V. longii, 1613-C rootstocks recorded more carbohydrates than the Salt Creek, 110-R and 140-Ru. Higher starch accumulation was recorded in SO4, Dogridge and 99-R compared to other rootstocks. The increase in the storage of carbohydrate in rootstock may help to increase the reserve food material in the vine required for developing bunch (sink).

The rootstocks 110-R, 99-R and Freedom rootstocks accumulated maximum amount of proteins in their leaves, while 1613-C, V. Champanii, 1103-P, Salt Creek accumulated low proteins in leaves. In shoots, higher proteins were recorded in Dogridge, SO4, 99-R compared to lowest in 1103-P, 140-Ru and 1613-C rootstocks. Though, in the present study, influence of these rootstocks on physiology and biochemistry of scion after planting, the inherent capacity of rootstock will still have the positive influence after grafting.

Principal component analysis

The principal component analysis (PCA) was performed on the biochemical composition from different parts of rootstocks are presented in Fig 1 and 2. PCA was used for visualization of the differences between rootstocks and biochemical composition in two dimensional space. Based on the correlation matrix, multivariate analysis was carried out to determine the relationship among the biochemical composition and grape rootstock. A clear differentiation were observed between parts of rootstocks and biochemical composition. The two principal components (PC) account for (97.48 %) of total variance of the data. The biochemicals such as, proteins, phenols, starch from shoots were positively contributed PC1 (93.57 %) and responsible for distribution of 110-R, Salt creek and Freedom rootstock. However, carbohydrates and starch from leaf and reducing sugar from root of rootstocks were negatively contributed to PC1 (3.916 %). Some biochemical from rootstocks situated in the positive part of both the principal components whereas some situated in negative part of principal components. Proteins composition from shoots are responsible to differentiate Dogridge and V. longii from other rootstocks which score for both the PCs.

According to the fig 1, PC1 and PC2 contributed 97.48 % of the total variance of data. The rootstocks 110-R and Freedom being differentiated by their high score for shoot carbohydrates in PC1 and PC2. 1613-C, V. longii, SO4 rootstocks showed similar pattern of their carbohydrates accumulation and values are positive for both PC1 and PC2. However, Dogridge and V. Champanii had similar patterns for accumulation of proteins from shoot values near to zero for PC1 and PC2. The protein and phenol composition of rootstocks are most important contributing factors for 140-Ru, 99-R, 1103-P, SO4 and V. Longii as in PC2.

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Table 1. Rootstocks and their species selected for the study

Sr. No.	Rootstocks	Percentage/ Species
1.	SO ₄	<i>V. berlandieri</i> x <i>V. riparia</i>
2.	1103 P	1 <i>Berlandierii</i> x <i>Rupestris</i>
3.	<i>V. Longii</i>	<i>V. longii</i>
4.	99R	99 R <i>Berlandierii</i> x <i>Rupestris</i>
5.	<i>V. champinii</i>	<i>Vitischampinii</i>
6.	Dog Ridge	<i>Vitischampinii</i>
7.	140 RU	<i>V. berlandieri</i> x <i>V. rupestris</i>
8.	St George	<i>Vitisrupestris</i>
9.	1613 C	<i>V. riparia</i> x <i>V. rupestris</i> x <i>V. vinifera</i> x <i>V. candicans</i> x <i>V. labruska</i>
10.	Freedom	<i>V.champinii</i> x (<i>V.solonis</i> x <i>V. othello</i>)
11.	Salt Creek	<i>Vitischampinii</i>
12.	110 R	<i>V. berlandieri</i> x <i>V. rupestris</i>



Table 2. Vegetative parameters in relation to different grape rootstocks

Rootstocks	Days to bud sprout	Shoot length (cm)	Shoot diameter (mm)	Inter nodal length (cm)	% rooting
SO ₄	10.50 ^b	35.00 ^f	5.320 ^{ab}	4.30 ^{cd}	79.00 ^{dc}
1103 P	9.00 ^d	39.00 ^{cd}	5.321 ^{ab}	4.20 ^{ed}	80.00 ^{abc}
V. Longii	9.00 ^d	42.00 ^b	5.300 ^{ab}	4.20 ^{ed}	82.00 ^a
99R	8.00 ^f	40.00 ^c	5.350 ^a	4.00 ^f	80.00 ^{abc}
V. Champinii	7.00 ^g	38.00 ^{de}	5.300 ^{ab}	3.50 ^g	79.60 ^{bc}
Dog Ridge	9.50 ^c	45.00 ^a	5.260 ^{ab}	4.10 ^{ef}	78.60 ^{dc}
140 RU	10.20 ^b	42.00 ^b	5.278 ^{ab}	4.40 ^{bc}	80.50 ^{abc}
St. George	8.20 ^{ef}	45.00 ^a	5.300 ^{ab}	4.30 ^{cd}	81.40 ^{ab}
1613 C	8.00 ^f	37.00 ^e	5.211 ^b	4.50 ^b	80.00 ^{abc}
Freedom	13.00 ^a	33.00 ^g	5.310 ^{ab}	4.80 ^a	79.00 ^{dc}
Salt Creek	9.00 ^d	35.00 ^f	5.225 ^{ab}	4.30 ^{cd}	82.00 ^a
110 R	8.50 ^e	30.00 ^h	5.228 ^{ab}	4.50 ^b	77.00 ^d
CV %	2.101	2.295	1.528	1.567	1.519
LSD 5 %	0.325	1.493	-	0.113	2.056
Significance	<.0001	<.0001	0.5709	<.0001	0.0018

Table 3. Biochemical Status in different parts of grape rootstocks.

Rootstocks	Carbohydrate (mg g ⁻¹)			Starch (mg g ⁻¹)			Protein (mg g ⁻¹)			Phenol (mg g ⁻¹)			Reducing Sugar (mg g ⁻¹)	
	Leaf	Shoot	Root	Leaf	Shoot	Root	Leaf	Shoot	Root	Leaf	Shoot	Root	Leaf	Shoot
SO ₄	117.54 ^c	368.8 ^{0^{gf}}	620.98 ^d	17.19 ^a	52.01 ^e	248.44 ^b	78.33 ^e	382.8 ^{0^b}	880.0 ^{0^{cd}}	154.0 ^{9^{ed}}	86.58 ^d	644.35 ^b	90.78 ^a	38.24 ^c
1103 P	95.85 ^e	300.2 ^{0ⁱ}	646.88 ^{dc}	10.94 ^d	81.47 ^d	216.74 ^c	70.67 ^f	262.1 ^{3ⁱ}	907.5 ^{0^{bc}}	133.9 ^{1^f}	46.93 ^e	729.57 ^a	63.00 ^d	31.47 ^e
V. Longii	88.88 ^f	310.0 ^{5ⁱ}	860.27 ^a	3.79 ^g	40.85 ^f	238.62 ^b	67.67 ^f	328.1 ^{3^e}	924.1 ^{7^b}	162.9 ^{6^{bc}}	44.15 ^{ef}	704.35 ^a	54.22 ^e	37.91 ^c
99R	100.04 ^d	320.3 ^{3^{ih}}	752.23 ^b	9.78 ^e	38.62 ^b	279.24 ^a	94.17 ^b	366.8 ^{0^c}	856.6 ^{7^d}	155.3 ^{0^{cd}}	101.5 ^{4^c}	613.04 ^c	67.56 ^c	55.80 ^a
V. Champinii	76.03 ^g	344.0 ^{5^{gh}}	624.55 ^d	12.19 ^c	122.5 ^{4^b}	113.62 ^g	44.67 ^h	334.4 ^{7^e}	784.1 ^{7^e}	72.70 ^{0^h}	32.50 ^g	552.17 ^d	30.44 ^f	30.24 ^e
Dog Ridge	138.88 ^a	412.9 ^{7^{de}}	640.12 ^{cd}	15.22 ^b	129.2 ^{4^a}	186.54 ^d	58.50 ^g	442.4 ^{7^a}	814.5 ^{2^e}	91.13 ^g	243.9 ^{7^a}	470.50 ^e	54.56 ^e	45.58 ^b
140 RU	87.99 ^f	389.4 ^{8^{ef}}	541.52 ^e	12.28 ^c	40.85 ^f	167.63 ^e	81.67 ^{de}	268.8 ^{0^{ih}}	974.1 ^{7^a}	192.5 ^{2^a}	28.50 ^g	646.09 ^b	74.22 ^b	27.91 ^f
St. George	62.46 ⁱ	432.5 ^{9^d}	642.41 ^{cd}	9.87 ^e	75.22 ^d	194.42 ^d	85.17 ^{cd}	289.4 ^{7^g}	565.0 ^{0^h}	146.9 ^{6^e}	31.80 ^f	417.39 ^f	53.44 ^e	33.58 ^d
1613 C	69.24 ^h	350.4 ^{7^g}	664.73 ^c	11.83 ^c	53.79 ^e	189.96 ^d	47.33 ^h	281.1 ^{3^{hg}}	593.3 ^{3^h}	90.78 ^g	34.76 ^f	408.70 ^f	60.22 ^d	37.91 ^d
Freedom	94.06 ^e	590.9 ^{8^b}	546.88 ^e	17.01 ^a	56.92 ^e	66.29 ⁱ	88.00 ^c	290.1 ^{3^g}	452.5 ^{0ⁱ}	169.9 ^{1^b}	102.0 ^{6^c}	375.65 ^g	69.00 ^c	34.47 ^d
Salt Creek	124.96 ^b	492.8 ^{7^c}	447.77 ^f	14.69 ^b	103.3 ^{5^c}	93.97 ^h	68.50 ^f	307.1 ^{3^f}	732.5 ^{0^f}	126.6 ^{1^f}	131.4 ^{5^b}	490.43 ^e	53.56 ^e	23.91 ^g



110 R	63.61 ⁱ	736.9 3 ^a	545.98 e	8.17 ^f	32.37 g	129.69 ^f	114.3 3 ^a	348.4 7 ^d	677.5 0 ^g	196.0 0 ^a	105.7 1 ^c	403.48 ^f	60.89 d	45.02 ^b
CV %	2.640	3.727	2.665	3.104	5.498	3.790	3.152	2.388	2.515	3.286	8.100	2.769	3.037	2.626
LSD 5 %	4.171	26.56 4	28.339	0.626	6.418	11.367	3.999	13.15 2	32.52 1	7.851	11.31 6	25.232	3.137	1.638
Significance	<.0001	<.000 1	<.0001	<.000 1	<.000 1	<.0001	<.000 1	<.000 1	<.000 1	<.000 1	<.000 1	<.0001	<.000 1	<.0001

Table 4. Effect of different grape rootstocks on gas exchange parameters and chlorophyll status

Rootstocks	Photosynthesis (μmol/mg/s)	Stomata Conductance (mm/s)	Transpiration rate (mmol H ₂ O m ⁻² s ⁻¹)	W.U.E. (μmol/mmol)	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)	Chlorophyll a and b (mg/g)	Total Chlorophyll (mg/g)
SO ₄	8.77 ^d	0.246 ^t	2.697 ^d	3.252 ^h	1.298 ^d	0.446 ^t	2.910 ^{gt}	1.893 ^t
1103 P	13.77 ^{cd}	0.341 ^d	3.195 ^d	4.310 ^{et}	1.797 ^c	0.503 ^{ed}	3.576 ^b	2.420 ^c
V. Longii	13.20 ^d	0.258 ^t	2.950 ^e	4.475 ^{ed}	1.101 ^h	0.388 ^g	2.838 ^g	1.653 ^g
99R	14.87 ^b	0.265 ^{et}	2.890 ^{et}	5.145 ^c	1.829 ^{bc}	0.514 ^{cd}	3.560 ^b	2.460 ^{bc}
V. Champinii	14.34 ^{bc}	0.403 ^{ab}	3.432 ^c	4.178 ^t	1.635 ^d	0.446 ^t	3.667 ^a	2.214 ^d
Dog Ridge	12.53 ^e	0.390 ^{bc}	3.700 ^a	3.386 ^{hg}	1.358 ^{gt}	0.440 ^t	3.085 ^e	1.944 ^t
140 RU	7.38 ^h	0.076 ^g	1.300 ^h	5.677 ^a	1.462 ^e	0.491 ^e	2.977 ^t	2.089 ^e
St George	14.84 ^b	0.264 ^{et}	2.724 ^g	5.448 ^b	1.840 ^{bc}	0.569 ^b	3.234 ^d	2.520 ^b
1613 C	13.98 ^c	0.279 ^e	2.804 ^g	4.986 ^c	1.781 ^c	0.522 ^c	3.408 ^c	2.422 ^c
Freedom	10.27 ^t	0.279 ^e	2.943 ^{et}	3.490 ^g	1.379 ^t	0.444 ^t	3.109 ^e	1.968 ^t
Salt Creek	16.07 ^a	0.421 ^a	3.590 ^{ab}	4.476 ^d	1.885 ^b	0.530 ^c	3.557 ^b	2.528 ^b
110 R	14.80 ^b	0.378 ^c	3.500 ^{bc}	4.229 ^t	2.115 ^a	0.612 ^a	3.454 ^c	2.820 ^a
CV %	2.655	3.614	2.794	2.210	2.222	1.943	1.538	1.995
LSD 5 %	0.580	0.018	0.140	0.165	0.061	0.016	0.085	0.075
Significance	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001	<.0001

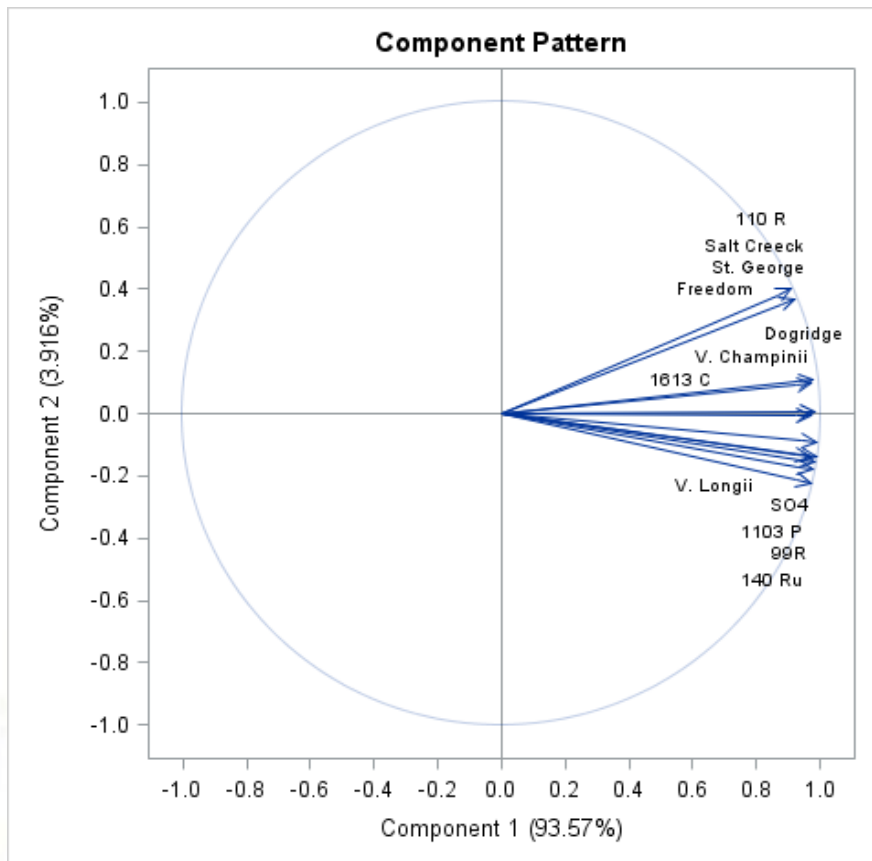


Fig 1: Rootstocks responsible for distribution of biochemical composition

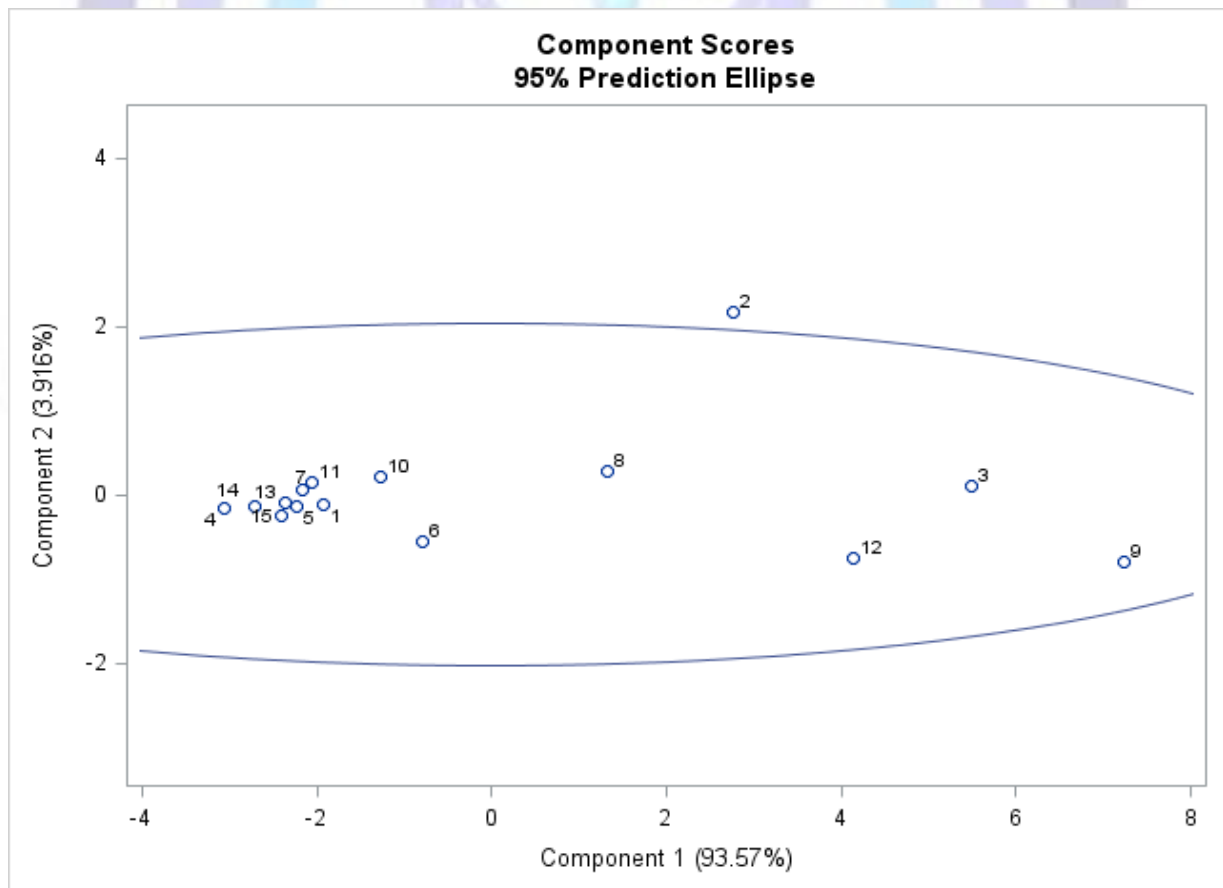


Fig 2: Differentiation of Rootstocks based on biochemical composition



Note: Number in the above figure indicates 1= Leaf carbohydrates;2=Cane carbohydrates;3=Root carbohydrates; 4=Leaf Starch; 5=Cane Starch, 6=Root Starch; 7=Leaf Proteins; 8=Cane Proteins; 9=Root Proteins; 10=Leaf Phenols; 11=Cane Phenols; 12= Root Phenols;13=Leaf ReducingSuagr; 14=Cane ReducingSugar; 15=Root ReducingSugar.

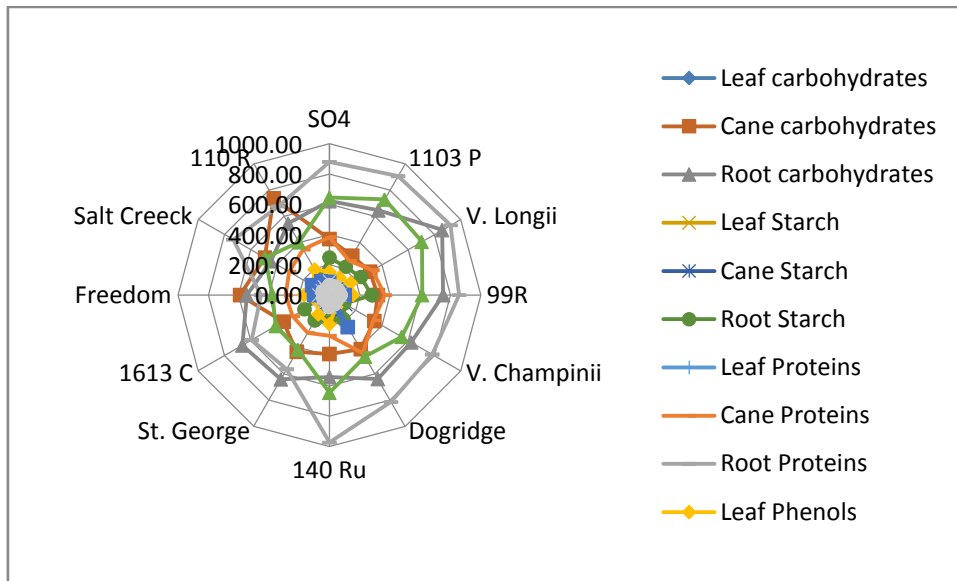


Fig 3: Accumulation of biochemical composition in different parts of rootstocks