



Changes in growth, photosynthetic activities, biochemical parameters and amino acid profile of Thompson Seedless grapes (*Vitis vinifera* L.)

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Abstract

The study on photosynthetic activity and biochemical parameters in Thompson Seedless grapes grafted on Dog Ridge rootstock and its impact on growth, yield and amino acid profile at various stages of berry development was conducted during the year 2012-2013. Leaf and berry samples from ten year old vines of Thompson Seedless were collected at different growth and berry developmental stages. The analysis showed difference in photosynthetic activity, biochemical parameters and amino acid status with the changes in berry development stage. Higher photosynthetic rate of $17.39 \mu\text{mol cm}^{-2} \text{s}^{-1}$ was recorded during 3-4mm berry size and the lowest ($10.08 \mu\text{mol cm}^{-2} \text{s}^{-1}$) was recorded during the veraison stage. The photosynthetic activity showed gradual decrease with the onset of harvest while the different biochemical parameters showed increase and decrease from one stage to another in both berry and leaves. Changes in photosynthetic activity and biochemical parameters thereby affected the growth, yield and amino acid content of the berry. Positive correlation of leaf area and photosynthetic rate was recorded during the period of study. Reducing sugar (352.25 mg g^{-1}) and total carbohydrate (132.52 mg g^{-1}) was more in berries as compared to leaf. Amino acid profile showed variations in different stages of berry development. Marked variations in photosynthetic as well as biochemical and amino acid content at various berry development stages was recorded and thereby its cumulative effect on the development of fruit quality.

Key words

Amino acid, Berry, Biochemical, Harvest, Photosynthesis, Veraison

Introduction

Grape is one of the major important fruit crops of the country, grown on an area of about 1,11,000 ha, with an annual production of 12,35,000 tonnes (Anonymous, 2012). Thompson seedless is a commercially accepted table grape variety in India both for domestic market and also for export. Earlier, vines growing on their own roots performed well since the soil and water condition were good. However, with the onset of problems like soil salinity, bad quality water and shortage of irrigation water due to drought resulted into reduced fruitfulness, thereby reducing yield of table grapes. The health of vine plays an important role in achieving higher fruit yield and better quality. In addition to irrigation, nutrition and sunlight, different means are followed by the grape growers to achieve the fruitfulness of vines. Among the

grape rootstock varieties, Dog Ridge (*Vitis champini*) is one of the most popular rootstocks with the grape growers, in addition to 110-R as the second alternative (Somkuwar *et al.*, 2006). Several studies have also shown that rootstocks influence various physiological parameters of the scions such as photosynthesis, stomatal conductance, transpiration rate, biochemical parameters such as total phenols, sugars, acid content, growth regulators (During *et al.*, 1997; Fernandez *et al.*, 1997; Nikolaou *et al.*, 2000; Iacono *et al.*, 1998); and morphological parameters (Satisha and Prakash, 2006), after grafting.

Grape berry is a fleshy fruit that grows in size and weight during the season, following an S shaped or double sigmoid curve pattern that can be divided into three different stages. After bloom (flowering) and fruit set, initial berry growth is associated with

rapid cell division and subsequent cell expansion. The phase of cell division (Stage I) is followed by a phase of cell expansion (Stage III) with an intermediate phase (Stage II) of a reduced growth called "lag-phase". Various growth regulators are also involved in controlling the berry growth of grapes and its chemical composition can be regulated by source-sink relationships (Kliwer and Dokoozlian, 2005). Assimilate supply from a source may be increased by increasing the leaf: fruit ratio, which generally leads to a larger fruit size in grapes (Petrie *et al.*, 2003). There are also variations in other phenological parameters like the time of bud burst, flowering and fruit set, fruit maturity and ripening. The cultural practices followed during the season play an important role in berry development and achievement of the quality grapes fitting into the required exportable standards. Considering this, an experiment was conducted to study growth, yield, photosynthesis, biochemical status and amino acid profile of Thompson Seedless grapes grafted on Dog Ridge rootstock.

Materials and Methods

The study was conducted during the fruiting season of 2012-2013 in the research and developmental plot of National Research Centre for Grapes, Pune, India. The soil of this region is black, having a slightly alkaline pH (8.0 to 8.5). Ten-year-old vines of 'Thompson Seedless' grafted on Dog Ridge rootstock were selected for study. The vines were planted at a spacing of 3 m between the rows and 1.8 m between the vines, accommodating about 1,800 vines per hectare. The row orientation was north-south. Irrigation and fertilizers were given as per the recommended schedule developed for this region. The plants were trained to flat roof gable system with four cordons in horizontal direction. The vines were pruned twice a year, once during March, after fruit harvest, which is referred to as foundation pruning mainly done to develop vegetative growth, and once during September-October, which is called forward pruning to encourage further bunch production. Forward pruning was performed in October, 2012 and the full bloom was attained at about 40-45 days after pruning.

About 100 g grape berries and leaf samples were regularly collected at 15-days interval from the fruit set stage until harvest and sample berries from bunches were collected. The samples were packed in polythene bags and brought to the laboratory in ice boxes. Sampling was done at four major berry development stages of 3-4mm berry size, 8-10mm berry size; veraison stage and harvest stage. The samples were washed thoroughly with distilled water, air-dried and stored at -20°C, prior to extraction and analysis. The berries were also collected at harvest to study the fruit quality. The mature berries were crushed in the mixer/grinder and the corresponding total soluble solids (°Brix) were recorded, using a hand-held refractometer, calibrated at a temperature of 20 °C (Model Erma Inc., Tokyo, Japan), before keeping the crushed samples in a -20°C deep freezer.

Growth parameters: The shoot and the internodal length (cm) and shoot diameter (mm) were recorded manually with the help of a measuring tape and a vernier calliper, however, leaf area (cm²) was measured using leaf area meter.

Photosynthetic activities: Photosynthetic rate, stomatal conductance and transpiration rate was recorded using Infra-Red Gas Analyser (IRGA) model (Li 6400, LI-COR Biosciences, Nebraska, USA). The observations were recorded during bright sunlight from 11.0 am to 12.30 pm.

Physico-chemical analysis of berry sample at harvest: Berry samples were collected during harvesting stage. These samples were crushed and the juice was analyzed by Foss analyzer method. The parameters analysed were total soluble solids (TSS), pH, total titratable acidity (TTA) and volatile acidity (VA).

Biochemical studies : The biochemical parameters were analysed in berries and leaves at different stages. The total carbohydrate and starch content were estimated by Anthrone method (Sadashivam and Manickam, 1996), reducing sugar by DNSA method (Sadashivam and Manickam, 1996), the total phenolic content using Folin-Ciocalteu method (Singleton and Rossi, 1965) and protein content (Lowry *et al.*, 1951). Total chlorophyll content of the leaves was estimated by DMSO method.

Amino acid analysis: The analysis of amino acids was performed using Perkin-Elmer Series 200 HPLC system (Perkin-Elmer India Limited, Mumbai, India) coupled to an API 2000 (ABS Sciex, Ontario, Canada) triple quadrupole mass spectrometer equipped with an electrospray ionisation (ESI) probe. The HPLC separation was carried out using an Atlantis dC₁₈ column (100x2.1mmx5µm) from Waters India Pvt Ltd., Bangalore. The mobile phase A composed of methanol:water (20:80) with 5mM ammonium formate and B composed of methanol: water (90:10) with 5mM ammonium formate. The column flow rate was 0.4ml min⁻¹ and column temperature 35°C. A gradient chromatography composed of 0-0.5 min 85% A, 0.5-6min 85-2% A, 6-11min 2% A, 11-12min 2-85% A and 12-20 min 85% A. The source parameters like nebulizer gas 40 psi, heater gas 60 psi, ion source temperature 550°C, ion spray voltage 5500V for positive polarity. All the standard solutions used under the study were stored at 4 °C.

The data were analyzed statistically using GLM procedure of SAS System software, version 9.3 (SAS,2004).

Results and Discussion

In general, the shoot length, shoot diameter, internodal length, leaf area varied significantly among different berry development stages (Table 1). However, the number of bunches did not change. Satisha *et al.* (2013), in their study reported that most of the vegetative growth parameters, such as average shoot

length, cane diameter and stock: scion ratio, were highest in Thompson seedless vines grafted on Dog Ridge rootstock. The various yield and quality parameters were recorded at the harvest stage. The berry length and berry diameter were recorded to be 17.92 mm and 14.58 mm respectively. Bica *et al.* (2000) observed a significant effect of rootstock on leaf area, chlorophyll content, stomatal conductance and quantum yield in Pinot Noir and Chardonnay grapevines. Increase in berry weight may also contribute to increase in average bunch weight. Yield per vine is result of bunch weight and number of bunches. Berries containing increased total soluble solids (above 20° Brix) are not preferred in the international market. However, in the local market grapes containing higher TSS is given due importance. Brix yield is said to be an important criterion for considering the yield level of vine (Somkuwar and Ramteke, 2006). Fruit composition parameters that eventually affect quality include soluble solids, organic acids, pH, phenolics and anthocyanins, monoterpenes and other components (Jackson and Lombard, 1993). The yield contributing parameters recorded during the harvest were the number of berries per bunch (110.0), 50-berry weight (120.96g), average bunch weight (250.67g) and yield per vine was 8.250kg. The quality parameters recorded were TSS (22.38 °C) and juice pH (3.59).

The rate of photosynthesis was higher during 3-4mm berry developmental stage (17.39 $\mu\text{mol cm}^{-2} \text{s}^{-1}$) which there after showed continuous decline (Table 2). Variation in photosynthetic activities in leaf was noticed in the present investigation. Increase in photosynthesis at berry setting stage (3-4 mm) might be due to the leaves that were young, hence more active and efficient photosynthetically. Possible reasons for general decrease in photosynthetic activity could be increase in total leaf area of canopy during the season, resulting in a decreased specific photosynthetic activity of the leaves; an increase in leaf age; as

well as a decreased demand for assimilates because of a decrease in actively growing vegetative sinks and berry growth. Stomatal conductance increased from 3-4 mm berry stage to 8-10 mm and was maximum at veraison stage (0.18 cm s^{-1}). However, there was a decrease in stomatal conductance at the harvest stage and the same trend was also observed for transpiration rate.

The chlorophyll content in leaf gives an indication of the efficiency of leaf to prepare food through photosynthesis. Significant differences were recorded in chlorophyll contents in leaf. The total chlorophyll content of leaf showed an increase from 3-4mm berry stage to veraison stage. However it reduced during the harvest stage. Increase in chlorophyll a was from 3-4 mm stage to harvest stage while chlorophyll b from 3-4 mm to veraison stage was noticed. The chlorophyll a:b ratio increased (Table 2) in leaves at the harvest stage (5.49).

Positive and negative correlations were found between vegetative and physiological parameters in Thompson Seedless grapevine grafted on Dog Ridge rootstock (Table 3). Among different parameters, shoot length was positively correlated with shoot diameter, leaf area and photosynthesis, however negative correlation was observed with stomatal conductance and transpiration rate. Similar results were also reported by Naor *et al.* (1997) in their experiment. Leaf area was positively correlated with photosynthesis while it was negatively correlated with stomatal conductance and transpiration rate. Increase in leaf area by increasing the number of shoots might have contributed to better photosynthesis (Somkuwar *et al.*, 2013). These results also supports the results obtained by Gao and Cahoon (1994) who reported that increase in leaves leads to a heavy canopy with increase in photosynthesis and stored carbohydrate in the plant system. The rate of photosynthesis also showed negative

Table 1: Vegetative parameter of Thomson Seedless vines at different developmental stages

Berry stages	Shoot length (cm)	Shoot dia (mm)	Inter nodal length (cm)	Leaf area (cm ²)	No. Bunches vine
3-4 mm	53.50	5.08	4.80	207.00	34.64
8-10 mm	54.00	5.10	4.82	210.00	34.64
Versions	55.00	5.12	4.85	211.00	34.64
At harvest	55.84	5.13	4.85	211.78	34.68

Table 2: Photosynthetic activities and chlorophyll content at different berry developmental stages

Stages	Photosynthesis ($\mu\text{mol cm}^{-2} \text{s}^{-1}$)	Stomatal conductance (cm s^{-1})	Transpiration rate ($\text{mmolH}_2\text{O m}^{-2} \text{s}^{-1}$)	Chl a (mg g^{-1})	Chl b (mg g^{-1})	Total Chl (mg g^{-1})	Chl a : Chl b ratio
3-4mm berry	17.39 ^a	0.14 ^c	3.12 ^b	1.12 ^d	0.44 ^a	1.56 ^c	2.54 ^d
8-10mm berry	10.20 ^d	0.15 ^b	3.20 ^b	1.45 ^c	0.45 ^a	1.90 ^b	3.25 ^c
Veraison	11.08 ^c	0.18 ^a	3.99 ^a	1.65 ^b	0.45 ^a	2.10 ^a	3.65 ^b
Harvest	11.51 ^b	0.06 ^d	2.55 ^c	1.76 ^a	0.32 ^b	2.08 ^a	5.49 ^a
CV%	1.558	3.472	2.721	2.123	2.223	2.092	1.733
LSD 5%	0.269	0.006	0.120	0.043	0.012	0.055	0.089
Sig.	** **	**	**	**	**	**	

correlation with stomatal conductance and transpiration rate. However, stomatal conductance was positively correlated with transpiration rate. Stomatal conductance has been reported to increase when fruit was present on grapevine (Poni *et al.*, 1994).

The data collected on various biochemical parameters in leaf and berries at different growth stages are presented in Table 4. Significant differences for reducing sugar content in Thompson Seedless grapes at various stages of berry development were recorded. Highest sugar content in leaf was recorded in 3-4mm stage (52.25 mg g^{-1}) while the concentration was reduced from 8-10mm berry development stage to the harvest. A rapid decrease in reducing sugar content at the harvest stage was recorded in leaf. In contrast to leaf, the concentration of sugar content in berries at different berry development stages was increased significantly from 3-4 mm berry stage (5.35 mg g^{-1}) to the harvest stage (352.25 mg g^{-1}). The reducing sugar content in berries of Thompson Seedless grapevine grafted on Dog Ridge rootstock at the harvest stage was found to increased many folds. The results of the present investigations are in confirmation with the results obtained by Gupta and Kaur (2000), who reported that the sugar concentration was increasing up to harvest but the storage of sugar converted to sucrose to glucose and fructose from 3-4 mm berry stage to harvest while more noticeable conversion of sugar happened between veraison to the harvest stage. However, Ali *et al.*, (2011) also reported a decrease in sugar concentration at the harvest stage. Grape berries act as a typical sink organ which rely on the use of available carbohydrate resource produced by

photosynthesis to supports their growth and development. In addition the number of factors assessing to monitoring of ripening, berry sugar concentration is one of the factors to monitor the maturity at harvest (Ali *et al.*, 2011). Any change in sugar concentration is mainly due to water loss from the berries at maturity. Our results of reduction of sugar at the time of harvest in leaf supports the findings of Ali *et al.* (2011) who also reported that any change in sugar concentration is mainly due to water loss.

Protein content in both leaf and berry showed significant changes in all the berry development stages. In leaf, protein content increased from 3-4 mm stage (13.49 mg g^{-1}) to veraison (22.73 mg g^{-1}) but then showed slight decline at the harvest stage (20.51 mg g^{-1}). Similarly, in berry, protein content increased from 3-4 mm berry stage (5.07 mg g^{-1}) to 8-10 mm berry stage (10.56 mg g^{-1}) while reduction of protein from 8-10mm till harvest (4.06 mg g^{-1}) was noticed in the berries (Table 5). The results of present investigation on reduction in protein content at the harvest stage are in accordance with the results of Marzia Giribaldi *et al.* (2007) who analyzed protein in grape berries by applying TCA/acetone based method and obtained more than $1 \text{ mg protein g}^{-1}$ berry fresh weight upto the sampling stage 3, while protein yield markedly and progressively decreased thereafter to reach about 0.3 mg g^{-1} berry fresh weight at the end of ripening. They also reported that after loading an equal amount of protein on the gel, the number of spots observed was higher before veraison and decreased thereafter. This explains that protein content in developing berries at green stages was much higher

Table 3 : Correlation studies between various vegetative parameters and photosynthetic activities

Pearson Correlation Coefficients, N = 4						
Prob> r under H0: Rho=0						
	Shoot length (cm)	Shoot diameter (mm)	Leaf area (cm ²)	Photosynthesis	Stomatal conductance	Transpiration rate
Shoot length (cm)	1	0.992*	0.989*	0.945*	-0.6605	-0.5292
Shoot diameter (mm)		1	0.974*	0.900*	-0.5648	-0.559
Leaf area (cm ²)			1	0.969*	-0.7217	-0.4034
Photosynthesis				1	-0.867	-0.3233
Stomatal conductance					1	0.00149
Transpiration rate						1

Table 4: Biochemical status of leaf and berries at different berry developmental stage of Thompson Seedless grapes grafted on Dog Ridge

Stages	Reducing sugar (mg g ⁻¹)		Protein (mg g ⁻¹)		Phenol (mg g ⁻¹)		Starch (mg g ⁻¹)		Total carbohydrate (mg g ⁻¹)	
	Leaf	Berry	Leaf	Berry	Leaf	Berry	Leaf	Berry	Leaf	Berry
3-4 mm Berry	52.25 ^a	5.35 ^d	13.49 ^c	5.07 ^b	3.89 ^c	2.62 ^b	5.01 ^c	4.61 ^b	45.62 ^c	27.98 ^d
8-10mm Berry	45.22 ^b	27.53 ^c	20.42 ^b	10.56 ^a	8.45 ^a	5.95 ^a	5.91 ^b	10.17 ^a	47.13 ^b	40.10 ^c
Veraison	14.92 ^c	125.10 ^b	22.73 ^a	3.83 ^c	8.45 ^a	1.89 ^c	6.80 ^a	1.38 ^d	31.44 ^d	45.08 ^b
At Harvest	3.04 ^d	352.25 ^a	20.51 ^b	4.06 ^c	7.51 ^b	1.97 ^c	4.89 ^c	3.09 ^c	48.97 ^a	132.52 ^a
CV %	3.591	4.258	2.512	3.039	2.807	3.399	2.537	4.15	0.937	2.189
LSD 5 %	1.428	7.484	0.667	0.246	0.273	0.145	0.197	0.275	0.559	1.852
Significances	**	**	**	**	**	**	**	**	**	**

whereas at maturity the concentration went down.

In general, phenolic content in leaf was more as compared to berries at all growth stages. Total phenol content in leaf increased significantly from 3-4mm (3.89 mg g⁻¹) to veraison (8.45 mg g⁻¹). Phenol increased nearly two-fold till veraison stage and then showed steep decrease at harvest. However in berry, phenols showed continuous increase upto 8-10 mm berry stage (5.95 mg g⁻¹) and then showed a sharp decrease in veraison (1.89 mg g⁻¹) and harvest (1.97 mg g⁻¹) stage (Table 5). Decrease in berry phenolic content could be attributed to the accumulation of water in berries during maturation.

Starch content in leaf increased from 3-4 mm berry stage (5.01 mg g⁻¹) to veraison (6.80 mg g⁻¹). Vegetative growth also stopped at 3-4mm berry size stage onwards. However, with the advancement of harvesting period there was decrease in starch content (4.89mg g⁻¹) as compared to the veraison stage. Almost a similar trend was noticed in grape berries in which starch content increased upto 8-10 mm berry stage (10.17 mg g⁻¹) and then sharply declined in veraison (1.38 mg g⁻¹) and harvest (3.09 mg g⁻¹). However, the extent of reduction in the starch content in berries was much higher as compared to leaf. The results obtained in the present study for starch content are in accordance with the results of Fischer and Ludders (1997). In their study on Cape gooseberry fruit, starch content increased more sharply during the first 10-20 days but then decreased steadily as the fruits developed and ripened. They also reported that leaf starch content was higher than fruits. Hu *et al.* (2009) while working on cucumber observed that the starch level was always higher in leaves than in the mesocarp tissues. Starch is generally regarded as the major storage carbohydrate in higher plants. The developing bunch as sink requires strong source during the berry development stage. The increased level of starch in leaves might explain that the source supply was sufficient for growing demand of the sink. There may be temporary carbohydrate reservoir in the stem that can support fruit development and maturation (Lapointe, 1998).

Total carbohydrate content in leaf and berries showed significant changes at different growth stages. In general, the total carbohydrate content increased from 3-4mm to 8-10 mm berry stage. Reduction in carbohydrate content from 8-10mm to veraison and again increase up to harvest was noticed for leaf.

Contrary to this, increase in starch content was observed in berry during the same stages. Maximum carbohydrate content was recorded at the harvest stage in both leaf (48.97 mg g⁻¹) and berry (132.52 mg g⁻¹). Increase in carbohydrate at the harvest stage might suggest that the transport from reservoir of stem to the developing bunch. Grape berries act as a typical sink organ depending on the use of available carbohydrate resource produced by photosynthesis to support their growth and development. From anthesis to veraison, imported carbon (in the form of sucrose) is almost equally partitioned between pericarp, seed growth and respiration. Carbon is mainly allocated to the pericarp and stored as hexoses, glucose and fructose (Ollat *et al.* 2000). These two sugars are the main carbohydrates of the mature berry pulp. Carbohydrates produced during photosynthesis are exported from the leaf as sucrose and transported in the phloem to the berry cluster (Davies and Robinson., 1996; Zhang *et al.*, (2006).

The data showed major differences in the amino acid accumulation pattern among different berry development stages, indicating the existence of wide range of variation (Table 5). High level of total amino acid at the time of harvest was recorded in Thompson Seedless vines. Most of the amino acids were present in very minute concentrations during the initial stages of berry development; however, arginine was maximum during 3-4 mm berry development stage and was then reduced till the harvest stage. At 3-4 mm berry stage, major amino acids present were alanine, arginine, proline, hydroxyproline, isoleucine, norleucine and serine, while in 8-10mm berry stage, aspartic acids, glutamic acid, glycine, lysine and ornithine were also found other than the amino acids present in 3-4 mm berry stage. With the onset of veraison, glycine, isoleucine, lysine, norleucine and serine were absent while at harvest stage, all the amino acids were present, except for aspartic acid, leucine, histidine and lysine. Among different amino acids, alanine, aspartic acid, histidine and ornithine showed gradual increase till veraison and then decrease at harvest stage, while proline, hydroxyproline, isoleucine, norleucine and serine showed decline till veraison and then increase at harvest stage. Leucine was completely absent in all the stages while arginine showed gradual decrease from 3-4mm berry stage upto harvest (Table 5). The amount of glutamic acid was found almost same at all the stages. Glycine increased

Table 5 : Amino acid profile (ppm) of Thompson Seedless grapes at different berry development stages

Stages	Ala	Arg	Asp	Glu	Pro	Gly	His	Hy.Pro	Isoleu	Leu	Lys	Nor	Orn	Ser
3-4 mm	0.149	9.04	0.00	0.00	0.262	0.00	0.00	0.122	0.020	0.0	0.00	0.202	0.00	0.588
8-10 mm	0.246	1.42	0.174	0.32	0.136	0.031	0.00	0.078	0.033	0.0	0.097	0.034	0.059	0.446
Veraison	0.482	1.03	0.308	0.384	0.097	0.00	0.376	0.011	0.00	0.0	0.00	0.00	0.094	0.00
Harvest	0.112	0.164	0.00	0.32	0.296	0.059	0.00	0.294	0.199	0.0	0.00	0.208	0.038	0.392

Ala=Alanine ,Arg=Arginine ,Asp=Aspartic acid, Glu=glutamic acid, Gly=Glycine, His=histidine, Hy.Pro=Hydroxy proline, Isoleu=isoleucine, Leu=leucine Lys=lysine, Nor leu=norleucine, Orn=ornithine, Ser=serine

upto 8-10mm berry stage while it was absent in veraison but again increased during harvest and lysine was found only at 8-10 mm berry stage. The changes in the concentration of amino acid at different berry development stages in a given variety might be due to the response to the cultural management practices during the season. Factors including cultivar, rootstock/scion combinations, vine nutrient management, vineyard site and growing season affect amino acid concentration within grapes (Bell and Henschke, 2005; Gump *et al*, 2002; Rodriguez-Lovelle and Gaudillere, 2002). The rate of increase was rather slow in the beginning, but accelerated after a lag phase (stage II) of berry development.

The concentration of alanine, glutamic acid, aspartic acid and arginine was highest as compared to other amino acids. The result obtained in this study confirms the results of Somkuwar *et al*, (2012) who reported that glutamic acid, aspartic acid and arginine were the major predominant amino acids. With the onset of veraison stage, alanine concentration in berry was found to be increased, however, at the harvest stage, alanine concentration was reduced. Stines *et al*. (2000) estimated the changes in free amino acids during berry development in four wine cultivars and reported that proline accumulation occurs during late ripening stages in all the cultivars tested, while that of arginine begins prior to veraison and stops at berry ripening. Arginine, in addition to serving as an important nitrogen reserve, participates in various physiological processes through nitric oxide pathway. A similar trend in accumulation of proline was observed in 'Thompson Seedless' grafted on Dog Ridge in the present study. Arginine is one of the major important amino acids present in 'Thompson Seedless' grapes (Hardie *et al.*, 1996) and its concentration increases from the middle of berry development stages but reduces later. Proline content was quite opposite and its concentration increased from a few days after veraison and there was a sudden surge during ripening of higher plants to water deficit and salinity stresses (Ashraf and Foolrad, 2007) and has numerous reviews over the last 20 years (Stines *et al*, 1999; Yang *et al*, 1999). It gets incorporated into purine and pyrimidine biosynthesis pathway in the form of Carbamyl-L-Aspartic acid. Free amino acids in grape berries were confined to mature berries was also reported by Spayd and Anderson-Bagge, (1996). In most of the plant species, either in the vegetative or reproductive tissues, proline and arginine metabolisms are linked through the amino acid ornithine (Verma and Zhang, 1999).

The study revealed that highest photosynthetic rate was during 3-4 mm berry stage and the lowest during veraison stage. The variation in content of biochemical parameters at different berry growth stages in leaf and berries was reported. Amino acid content also varied in different berry development stages.

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