



# Leaf Area Index, Photosynthesis and Chlorophyll Content Influences Yield and Quality of Nanasaheb Purple Seedless Grapes under Semi-arid Condition

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## Authors' contributions

This work was carried out in collaboration among all authors. Author RGS conceptualized the present experiment and implementation of research. Author PBK prepared the manuscript and performed the analytic calculations and numerical simulations. Authors ASJ and PKA collected and arranged the data. Authors PHN and NAD edited the manuscript. All authors read and approved the final manuscript.

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## ABSTRACT

Canopy management practices in grapes are crucial for achieving high-quality production. The study was conducted with different treatments of number of leaves above the bunch (10, 12, 14, 16, and >16 leaves). Significant variation was recorded on photosynthetic activity, yield and quality of Nanasaheb Purple Seedless grape variety. The increase in leaf number enhanced total leaf area and photosynthetic capacity while, it had negative impact on bunch weight, berry size and total soluble solids (TSS). Retaining 10 leaves above bunch with leaf area of 1980.00 cm<sup>2</sup> /bunch resulted in maximum bunch weight (450.20 g), 50-berry weight (250.13 g) and yield/vine (13.60 kg) while, minimum bunch weight (400.00 g), 50-berry weight (222.30 g) and yield/vine (11.22 kg) were observed by retaining >16 leaves above the bunch. However, retaining 10 leaves above the bunch on a shoot with leaf area/shoot (2475.00 cm<sup>2</sup>), leaf area/vine (59400.00 cm<sup>2</sup>), leaf area/bunch (1980.00 cm<sup>2</sup>) and leaf area/g berry weight (4.36 cm<sup>2</sup>/g) were sufficient to obtain good quality production in Nanasaheb Purple Seedless grape variety spaced at 9 feet X 5 feet distance.

*Keywords: Leaf area; chlorophyll content; photosynthetic activity; yield; quality.*

## 1. INTRODUCTION

For successful grape production, canopy management plays a crucial role, which involve training systems that tailored according to the soil type in a particular region, vine vigor and weather conditions that help to expose maximum leaf area to sunlight lead to increase yield potential [1]. In viticulture, balancing leaf area and fruit is critical for accomplishing desired fruit composition. This balance is achieved through canopy management practices which include techniques to adjust the number of leaves, shoots and fruits, therefore producing ideal canopy microclimate. The purpose of canopy modifications is to enhance production potential reduce disease and pest incidence and facilitate mechanization [1]. The aim of these practices is to produce high-quality grapes with increased sugar levels [2]. Furthermore, canopy management helps to maintain the vine's source (leaves): sink (bunch) balance [3]. Slight changes in canopy microclimates influenced by alteration in leaf areas can shorten the grape maturation period [4]. Falling leaf area can also augment nutrient uptake which helps in grape ripening and enhancing photosynthetic activity of the remaining leaves [5]. Leaf removal in the maturing stage is common in high-vigor and trained vine canopies [6]. Research indicated proper and timely leaf removal can lead to early maturation or delay maturity of grapes [7,8]. Fruit exposure to direct sunlight increases its temperature, aiding in malic acid degradation and improving the sugar-acid ratio [7]. In tropical region, grape varieties can achieve sufficient soluble solids required for quality grape production but this is not applicable in case of color development [9]. Sugar enzyme activity

more in the temperature range of 8 to 33°C differ from those color enzyme activity which required 17 to 26°C temperature [9,10]. Temperatures above 30°C can inhibit anthocyanin synthesis after the veraison stage. Research on ecophysiology has established the leaf area to fruit ratio as a key viticultural index to define a well-balanced vineyard capable of producing high-quality grapes [11]. The optimal leaf area to fruit ratio is between 0.8 and 1.2 m<sup>2</sup>/kg to achieve early maturity and quality [11]. Most of the previous studies have focused on determining the leaf area per unit weight of fruit to maximize sugar concentration a common indicator of berry ripeness. The optimum leaf area for color development in grapes varies depending on the grape cultivar and specific vineyard conditions. Auzmendi and Holzapfel [12] reported that the leaf area to fruit weight ratio (ranging from 6 to 12 cm<sup>2</sup>/g for fresh weight, 8 to 10 cm<sup>2</sup>/g for total soluble solids concentration and 9 to 12 cm<sup>2</sup>/g for anthocyanin content) plays a crucial role in berry composition and color development. To investigate the actual leaf area required to produce quality grapes with good color for export, this study aims to understand the effect of leaf retention above the bunch on leaf area, leaf area index, yield and quality of Nanasaheb Purple Seedless grapes grafted on Dogridge rootstock under semi-arid conditions.

## 2. MATERIALS AND METHODS

The study was conducted during 2023-24 at ICAR-National Research Centre for Grapes, Pune. The experimental site located in mid-west Maharashtra at an altitude of 559 meters above MSL (18.32°N, 73.51°E). Nanasaheb Purple Seedless grapevines, grafted on Dogridge

rootstocks were planted at a spacing of 9 feet x 5 feet and trained to extended Y- trellis maintaining 0.5 cane/feet<sup>2</sup> (24 canes/vine) on each vine. All the recommended standard cultural practices were followed to maintain healthy vine during the period of study. Five treatments with variation in leaf number above the bunch were evaluated as 10, 12, 14, 16 and >16 leaves, with each treatment replicated five times. The experiment was conducted in Randomized Block Design (RBD). LAI and PAR were recorded using LaiPen LP 110 device. LAI was calculated as leaf area per ground area (m<sup>2</sup>/m<sup>2</sup>), and PAR measured in  $\mu\text{mol photons m}^{-2}\text{s}^{-1}$ , quantified the photosynthetic photon flux density (PPFD). Assimilation rate, stomatal conductance, intercellular CO<sub>2</sub> and transpiration rate were measured using an Infra-Red Gas Analyzer (IRGA model Li 6400, LI-COR Biosciences, NE, USA) on fifth to sixth matured leaves from the shoot tip, between 11 am and 12:30 pm. Leaf area was determined using the linear method (LBK method) with the formula: Leaf area (A) = L x B x K (0.810) and expressed in cm<sup>2</sup>. Total leaf area/shoot, per vine and per bunch was calculated by multiplying the leaf area of individual leaf by the number of leaves per shoot, shoots per vine and dividing by the number of bunches per vine, respectively. Average bunch weight was derived from the mean weight of five randomly selected healthy bunches per replication while, the average weight of 50 berries was calculated and expressed in grams. The number of berries per bunch was averaged from five bunches per treatment. After-maturity, grapes from five vines in each treatment were harvested and weighed to calculate average yield/vine and was expressed in kilograms. Total soluble solids (TSS) were measured with a portable handheld refractometer (Erma Refractometer, Japan) at room temperature and total acidity (TA) was determined using OenoFoss (FTIR based wine analyzer) and expressed in g/l. Chlorophyll content was estimated by Arnon's (1949) method.

## 2.1 Statistical Analysis

The research trial laid out in randomised block design with five treatment and five replications. The data analysis was performed using analysis of variance (ANOVA) following the methodology outlined by Panse and Sukhatme (1995).

## 3. RESULTS AND DISCUSSION

The data recorded on effect of the number of leaves above the bunch on various leaf area

parameters are presented in Table 1. The maximum individual leaf area was recorded in 10 leaves above the bunch (165.00 cm<sup>2</sup>) treatment which was at par with 12 leaves above the bunch (162.10 cm<sup>2</sup>) while, minimum leaf area was recorded in > 16 leaves above the bunch (150.50 cm<sup>2</sup>) treatment. As the number of leaves above the bunch increased (from 10 to > 16), the leaf area per shoot, per vine and per bunch was also significantly increased. The leaf area/shoot increased from 2475.00 cm<sup>2</sup> (10 leaves above the bunch) to 3762.5 cm<sup>2</sup> (>16 leaves above the bunch). Similarly, the leaf area per vine increased from 59400.00 cm<sup>2</sup> to 90300.00 cm<sup>2</sup> while, the leaf area per bunch increased from 1980.00 cm<sup>2</sup> to 3225.00 cm<sup>2</sup>. The leaf area per gram of berry weight also increased from 4.36 cm<sup>2</sup>/g to 8.06 cm<sup>2</sup>/g. The optimum leaf number enhanced the overall leaf area, potentially contributing to higher photosynthetic capacity (source) and resource distribution (sink) for grape development. Similar conclusions were also reported by Thoke et al. [13]. Cataldo et al. [14] reported maximum leaf area in the control (1.62±0.34) treatment and lowest in the four-leaf removal (1.19±0.43) treatment. The outcome of the present study is in agreement with the findings of Candor et al. [4], Somkuwar et al. [15], Somkuwar et al. [1]. Somkuwar et al. [16,17] suggested that maintaining 12 leaves above the bunch with 63820.80 cm<sup>2</sup> in Crimson Seedless and 14 leaf above the bunch with 69312.00 cm<sup>2</sup> leaf area per vine Manjari Kishmish were sufficient for higher yield and better-quality grapes.

Bunch characteristics and yield/vine as influenced by number of leaves above the bunch are presented in Table 2. The average bunch weight decreased from 450.20 g (10 leaves above the bunch) to 400.10 g (>16 leaves above the bunch). The same trend was also observed for berry diameter as it decreased from 21.20 mm to 18.30 mm. The number of bunches/vines remained relatively stable, with slight variations while, the number of berries/bunch indicated non-significant difference across treatments. The 50-berry weight decreased from 250.13 g to 222.30 g and the yield/vine also decreased from 13.60 kg to 11.22 kg. The total soluble solids (TSS) decreased from 19.20 °Brix in 10 leaves above the bunch to 17.40 °Brix in >16 leaves above the bunch. However, the acidity levels remained relatively stable, with slight variations. The higher leaf numbers might have reduced TSS indicating a potential dilution of sugar content in grape berries which could affect

sweetness and overall berry quality. The increase in leaf number which enhanced the total leaf area but it did not improve bunch characteristics and yield. In fact, an excessive number of leaves might have negatively impacted both berry quality and yield. Potential of a vine to produce carbohydrates necessary for fruit production and vegetative growth depend on its actual leaf area [1]. Somkuwar et al. [16,17] reported that maintaining 12 leaves above the bunch in Crimson Seedless and 14 leaves above the bunch in Manjari Kishmish lead to enhance the productivity and raisin quality. However, excessive leaf retention decreased yield and quality. According to Somkuwar et al. [1] an optimum leaf area maximizes the rate of photosynthesis which helps meet the carbohydrate demands of the fruit (bunch). Additionally, Somkuwar et al. [18] reported 10.09 kg yield/vine in the control group and 12.75 kg in the shoot thinning treatment at the 6-7 leaf stage. Similar results were also reported by Somkuwar et al. [15] and Candar et al. [4].

The effect of varying leaf number on leaf area index (LAI) and photosynthetically active radiation (PAR) are presented in Fig. 1. LAI increased from 1.42 in 10 leaves above the bunch to 2.16 in >16 leaves above the bunch demonstrating widespread leaf coverage. However, PAR decreased from 0.152  $\mu\text{mol photon m}^{-2} \text{s}^{-1}$  to 0.098  $\mu\text{mol photon m}^{-2} \text{s}^{-1}$ .

Higher leaf cover increases shading and reduces light penetration in the canopy, potentially affecting photosynthesis. Somkuwar et al. [16,17] reported leaf area index (LAI) of Crimson Seedless and Manjari Kishmish increased with the number of leaves above the bunch (1.33 to 2.02 and 1.42 to 1.66  $\text{m}^2/\text{m}^2$  respectively) while, PAR value reduced as number of leaves increased from 0.59 to 0.41 and 0.14 to 0.94  $\mu\text{mol photon m}^{-2} \text{s}^{-1}$  respectively. Thoke et al. [13] also reported that the number of leaves per shoots and per vine increased, potentially leaf count. The leaf area and LAI also increased because of increase in number of leaves per vine which contributes to elevated LAI. Burg et al. [19] reported increased LAI from 1.86 to 2.22  $\text{m}^2/\text{m}^2$  in nine different grape varieties. The result of the present study suggests that an increase in leaf density positively correlates with a higher leaf area/ground area. Poni et al. [6] observed that due to defoliation of leaf (removed about 70% of the shoot leaf area) net carbon exchange rate (NCER) per vine significantly decreased. This reduction in NCER was linked to a decrease in PAR reaching the vine canopy. Optimizing leaf density to strike a balance between maximizing leaf area for efficient light interception and minimizing shading effects is essential for optimizing crop yield and resource utilization. Our study also aligns with the finding of Kang et al. [20], Junges et al. [21], Burg et al. [19], Somkuwar et al. [15].

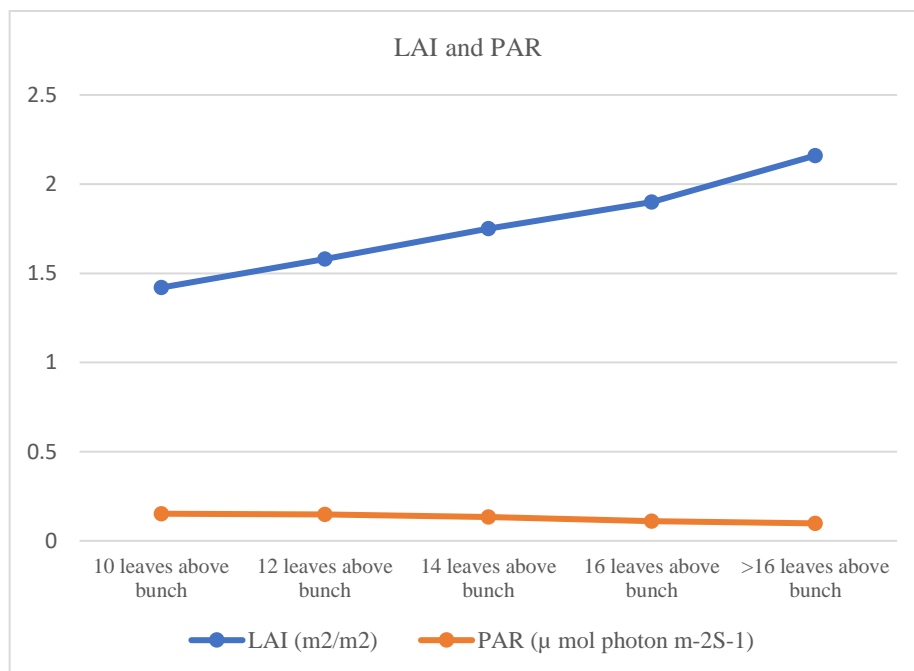


Fig. 1. Effect of leaves on leaf area index and PAR of vine in Nanasahab Purple variety

**Table 1. Effect of leaves on total leaf area in Nanasaheb Purple variety**

Leaf above the bunch	Leaf area/leaf (cm <sup>2</sup> )	Leaf area/shoot (cm <sup>2</sup> )	Leaf area/vine (cm <sup>2</sup> )	Leaf area/bunch (cm <sup>2</sup> )	Leaf area/g berry wt. (cm <sup>2</sup> /g)
10 leaves above bunch	165.00	2475.00	59,400.00	1980.00	4.36
12 leaves above bunch	162.10	2755.70	66,137.00	2204.60	5.12
14 leaves above bunch	160.50	3049.50	73,190.00	2559.10	6.17
16 leaves above bunch	157.80	3313.80	79,531.50	2840.39	6.92
>16 leaf above bunch	150.50	3762.5	90,300.00	3225.00	8.06
S Em ±	1.09	27.42	658.2	24.00	0.06
CD at 5 %	3.29	82.22	1973.5	71.95	0.18

**Table 2. Effect of leaves on bunch characters and yield in Nanasaheb Purple variety**

Leaf above the bunch	Av. bunch weight (g)	Berry diameter (mm)	No of bunches/ vine	No of berries/ bunch	50-berry wt. (g)	Yield/vine (kg)	TSS (°Brix)	Acidity (g/L)
10 leaves above bunch	450.20	21.20	30.20	90.00	250.13	13.60	19.20	5.25
12 leaves above bunch	430.25	20.40	30.00	89.50	240.36	12.90	18.80	5.20
14 leaves above bunch	415.35	20.00	28.60	90.20	230.25	11.87	18.20	5.30
16 leaves above bunch	410.30	19.50	28.00	90.00	228.00	11.48	17.20	5.25
>16 leaves above bunch	400.10	18.30	28.00	90.00	222.30	11.22	17.40	5.10
S Em ±	2.82	0.13	0.19	NS	1.56	0.08	0.12	0.03
CD at 5 %	8.45	0.40	0.5	NS	4.68	0.24	0.38	0.11

**Table 3. Effect of leaves on photosynthetic activities in Nanasaheb Purple variety**

Leaf above the bunch	Assimilation rate (µmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> )	Stomatal conductance (mmol CO <sub>2</sub> m <sup>-2</sup> s <sup>-1</sup> )	Intercellular CO <sub>2</sub> (Ci) (ppm)	Transpiration rate (mmol H <sub>2</sub> O m <sup>-2</sup> s <sup>-1</sup> )
10 leaves above bunch	13.10	0.12	245.50	2.56
leaves above bunch	12.85	0.13	248.15	2.51
14 leaves above bunch	12.55	0.14	245.11	2.53
16 leaves above bunch	12.15	0.13	243.22	2.53
>16 leaves above bunch	10.37	0.12	241.26	2.52
S Em ±	0.08	0.001	NS	NS
CD at 5 %	0.25	0.003	NS	NS

**Table 4. Effect of leaves above bunch on chlorophyll content in leaves of Nansah Purple variety**

Leaf above the bunch	Chlorophyll a (mg/ml)	Chlorophyll b (mg/ml)	Total chlorophyll (mg/ml)
10 leaves above bunch	17.50	3.85	21.35
12 leaves above bunch	17.15	3.67	20.82
14 leaves above bunch	16.35	3.65	20.00
16 leaves above bunch	16.20	3.53	19.73
>16 leaves above bunch	15.64	3.50	19.14
SEm ±	0.01	0.02	0.13
CD at 5 %	0.33	0.07	0.40

**Table 5. Correlation coefficients between different growth and yield parameters as influenced by number of leaves maintained above the bunch**

parameters	Leaf area index (m <sup>2</sup> /m <sup>2</sup> )	PAR (μ mol photon m <sup>-2</sup> S <sup>-1</sup> )	Leaf area/vine (cm <sup>2</sup> )	Leaf area/bunch (cm <sup>2</sup> )	Leaf area/gram of berry wt. (cm <sup>2</sup> /g)	Total chlorophyll content (mg/ml)	Av. bunch wt. (g)	Yield/vine (kg)
Leaf area index (m <sup>2</sup> /m <sup>2</sup> )	1							
PAR (μ mol photon m <sup>-2</sup> S <sup>-1</sup> )	-0.974	1						
Leaf area/vine (cm <sup>2</sup> )	0.884	-0.952	1					
Leaf area/bunch (cm <sup>2</sup> )	0.998	-0.983	0.892	1				
Leaf area/gram of berry wt. (cm <sup>2</sup> /g)	0.998	-0.978	0.880	0.999	1			
Total chlorophyll content (mg/ml)	-0.984	0.956	-0.827	-0.989	-0.992	1		
Av. bunch wt. (g)	-0.958	0.912	-0.771	-0.960	-0.968	0.986	1	
Yield/vine (kg)	-0.949	0.933	-0.797	-0.961	-0.966	0.987	0.988	1

Photosynthetic parameters resulted into decrease in assimilation rate from 13.10  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in 10 leaves above the bunch to 10.37  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in >16 leaves above the bunch. Slight variation was observed in stomatal conductance. It was increased from 14 leaves above the bunch (0.14) to 16 leaves above the bunch (0.13) while the stomatal conductance was again decreased in >16 leaves above the bunch (0.12) treatment. However, intercellular  $\text{CO}_2$  and transpiration rate was remained relatively stable. The decrease in the assimilation rate in a greater number of leaves was likely because, beyond a certain point, additional leaves may not contribute effectively to photosynthesis, possibly due to increased shading and reduced light penetration. Possible reason for general decrease in photosynthetic activity could be the increase in total leaf area of canopy [18]. The result of the present study aligns with the findings of our earlier results [16,17] where least variation in photosynthetic activities in Crimson Seedless and Manjari Kishmish variety was recorded after creating different variations of leaves above the bunch. Cataldo et al. [14] also reported non-significant result in leaf gas exchange parameters between four leaf removal and eight leaf removal during two-year study.

The efficiency of leaf to prepare food through photosynthesis depends upon chlorophyll content of leaf. The decrease in leaf chlorophyll was recorded from 17.50 mg/ml in 10 leaves above the bunch to 15.64 mg/ml in >16 leaves above the bunch (Table 4). Chlorophyll b also decreased from 3.85 mg/ml to 3.50 mg/ml, However, total chlorophyll content in leaf of grapevine also recorded similar trends of decrease from 21.35 mg/ml to 19.14 mg/ml. Thus, the decrease in chlorophyll content in more leaves indicated reduced chlorophyll synthesis, or increased degradation, potentially affecting photosynthetic efficiency. Petrie et al. [22] reported that leaf removal resulted in an increase in or retention of chlorophyll which also occurred for the full leaf removal crop treatment. Somkuwar et al. [16,17] reported that beyond optimum leaf number above the bunch (12 and 14 leaves respectively), chlorophyll content per leaf begin to decrease in Crimson Seedless and Manjari Kishmish grape variety.

Both positive and negative correlations were observed between the growth and yield parameters in Nanasaheb Purple Seedless grapes (Table 5). Leaf area index was positively

correlated with leaf area/vine (0.884), leaf area/bunch (0.998) and leaf area/gram of berry wt. (0.998) highlighting the influence of leaf density on total leaf area. However, the PAR values showed a strong negative correlation in leaf area index (-0.974), increased leaf density reduced light penetration.

#### 4. CONCLUSION

Optimum leaf area management per vine in any grape variety is an important parameter for enhancing the yield and quality. Retaining 10-12 leaves above the bunch in Nanasaheb Purple Seedless grape variety provides the best balance, maximizing leaf area and photosynthetic capacity while maintaining desirable bunch weight, berry size, yield/vine and total soluble solids (TSS). Excessive leaf retention (>16 leaves above the bunch) negatively impacts these quality parameters. Therefore, precise canopy management practices with an optimum leaf area/shoot (2475.00  $\text{cm}^2$ ), leaf area/vine (59400.00  $\text{cm}^2$ ), leaf area/bunch (1980.00  $\text{cm}^2$ ) and leaf area/g berry weight (4.36  $\text{cm}^2/\text{g}$ ) can be achieved by retaining 10 leaves above the bunch for producing high-quality grapes.

#### DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

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#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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