iVES OENO One vine & wine

DOI: https://doi.org/10.20870/oeno-one.2024.58.1.7827

OPEN ACCESS

ORIGINAL RESEARCH ARTICLE

Vine spacing of *Vitis vinifera* cv. Shiraz/101-14 Mgt. I. Root growth and Physiological activity

J.J. Hunter and C.G. Volschenk

[1] ARC Infruitec-Nietvoorbij, Private Bag X5026, Stellenbosch, South Africa

*correspondence: hunterk@arc.agric.za

Associate editor: Vivian Zufferey

Received: 17 October 2023

Accepted: 14 December 2023

Published: 24 January 2024

This article is published under the **Creative Commons licence** (CC BY 4.0).

Use of all or part of the content of this article must mention the authors, the year of publication, the title, the name of the journal, the volume, the pages and the DOI in compliance with the information given above.

ABSTRACT

Knowledge of vine reaction to plant spacing under relatively high potential soil conditions is limited. This study comprised effects of vine spacing (with fixed row spacing) of Shiraz (clone SH 9C)/101-14 Mgt on a high potential soil in the Breede River Valley, Robertson, South Africa, on soil conditions, root system distribution, and vine physiological reaction. The vineyard was planted in 2008 to a VSP trellis, with a fixed row spacing of 2.2 m and a row orientation of approximately NNE–SSW (30°). In-row vine spacing changed from 0.3–4.5 m with increments of 30 cm (from 15151–1010 vines/ha), totalling 15 treatments. After the completion of cordon development, results were generated over six seasons. Soil conditions seemed generally uniform and showed no obvious characteristics that could have affected treatments differentially. An increase in density occurred in fine, extension and permanent root categories with closer spacing, increasing the total number of roots/ha. Roots penetrated deeper with closer vine spacing. Vine spacing impacted physiological parameters and revealed a complex interplay between root distribution, vine structure expansion, canopy microclimate, water relations, photosynthetic output, berry temperature, carbon distribution, and day/night recovery that would affect grape composition and wine quality. Optimum vine spacing appeared to be 1.8 m, both closer and wider spacing leading to limitations in physiological activity. Interactions amongst below- and aboveground growth and physiological parameters are comprehensively discussed with a focus on sustainability.

KEYWORDS: Vine spacing, roots, physiology, canopy, microclimate, water, sustainability

INTRODUCTION

Plant spacing choices during establishment of new vineyards are generally dictated by soil physical and chemical properties (determining drainage, water holding capacity and mineral yielding potential) as well as the scion–rootstock combination (Richards, 1983; Hunter *et al*., 2010; Hunter *et al*., 2016a). Given the relevant climate change scenarios, such as drought, high temperature, heat waves and high vapour pressure deficits, that may affect grape growing regions globally, sustainable soil management as well as drought and high temperature tolerant genotypes (scions and rootstocks) have become even more important considerations than before (Southey, 1992; Medrano *et al*., 2015; van Leeuwen and Destrac-Irvine, 2017; Santos *et al*., 2020). Sustainability at the farm level seems to shift more and more towards an emphasis on production per water availability/use per unit surface area. It more often leans towards natural soil water holding capacity and favourable/bearable ambient temperature conditions as determining factors, owing to the increasing scarcity/unsuitability of irrigation water and changes in climatic conditions. This complicates production, quality objectives, and strategies at a practical level.

Potential root development for maximum soil colonisation seemed to be a key consideration in grapevine establishment to buffer adverse climatic conditions and promote stability in growth and production from season to season (Champagnol, 1979; Richards, 1983; Archer and Strauss, 1985; Hunter, 1998a; Hunter *et al*., 2016a; Archer and Saayman, 2018; van Leeuwen *et al*., 2019). Richards (1983) indicated soil temperature, water holding capacity, aeration, nutrition, and physical properties, such as the presence of restrictive (e.g., compact/dense, acid, saline) soil layers, as factors dictating the extent of grapevine root distribution and activity. Despite differences between so-called warm versus cold viticulture regions, the seasonal pattern of root growth activity is reasonably well known (Van Zyl, 1984; Mohr, 1996), but root mass/penetration/distribution and associated physiological responses may be affected by *inter alia*, scion–rootstock selection, specifically the rootstock (Nagarajah, 1987; Swanepoel and Southey, 1989), plant density (Champagnol, 1979; Archer and Strauss, 1985; Hunter, 1998a; van Leeuwen *et al*., 2019), trellising (Archer *et al*., 1988), soil texture (Nagarajah, 1987), tillage and mulching (Van Huyssteen and Weber, 1980; Saayman, 1982; Richards, 1983), cover crops (Linares Torres *et al*., 2018), soil pH alteration (Conradie, 1983), irrigation (Richards, 1983; Van Zyl, 1984), pruning levels (Slavtcheva and Pourtchev, 2003), canopy manipulation/partial defoliation (Hunter and Le Roux, 1992; Hunter *et al*., 1995b), and crop load (Edson *et al.*, 1995). Under dryland conditions on a medium potential, depth-restricted soil, the higher root densities of narrowly spaced vines caused a rapid seasonal depletion of soil water in all soil layers, with earlier stomatal closure and a reduced transpiration rate (Archer and Strauss, 1989). Under supplementary irrigation conditions and with similar root growth responses, both leaf and bunch water potential decreased, and abscisic acid concentrations in the leaf xylem sap increased with narrower spacing (Hunter, 1998b). Plant spacing affected canopy exposure and other microclimatic parameters, with wider spacing favouring photosynthetic activity. The physiological effects resulted in grapes ripening under higher water deficit conditions but with improved quality under both dryland and supplementary irrigation when vines were narrowly spaced.

Each soil profile and texture within general classifications, either more or less favourable for plant growth, would require a unique cultivation approach. Irrespectively, in all studies, root distribution and penetrable depth seemed a critical, common denominator for vigour and yield (Hunter *et al*., 2016a; Archer and Saayman, 2018). Although similarities in vine response to different cultivation conditions surfaced in studies, systematic results on the reaction of the vine in high potential, deep soils remained unresolved.

This study aimed to clarify the sole effect of vine spacing (with fixed row spacing) on root system distribution and physiological reaction under relatively high potential soil conditions and irrigation. The study targeted physiological responses and vineyard sustainability and longevity.

MATERIALS AND METHODS

1. Experiment layout and practices

Vitis vinifera cv. Shiraz (clone SH 9C)/101-14 Mgt was planted during the spring of 2008 with an approximate NNE-SSW (30°) row orientation at the Robertson experiment farm of ARC Infruitec-Nietvoorbij, located in the Breede River Valley, South Africa (GPS coordinates 33°49'29.87"S; 19°52'50.67"E). Vines were spaced from 0.3–4.5 m with incremental increases of 30 cm from narrowest to widest, representing 15 treatments (15,000–1000 vines/ha). Row spacing was fixed at 2.2 m. Vines were double-cordon-trained on a VSP trellis with four sets of movable foliage wires. Each treatment was replicated four times with two buffer rows per treatment on a total surface of approximately 0.9 ha. Measurements were done in the centre row of the replicate. Irrigation was applied according to crop factors (that changed from 0.3–0.6 as the season progressed). An average daily ET_0 of 5.0–6.7 mm was recorded, and the volume of irrigation water was applied at 10–28 mm/week, excluding erratic summer rain. To enhance the complete cordon development of all treatments, vines were pruned to one-bud spurs for two winters (2010 and 2011) after planting, where after two-bud spurs with 15 cm inter-spur spacing were pruned since the winter of 2012. Spurs were spaced equally, irrespective of treatment. The number of spurs increased according to cordon length. Vines were irrigated and fertilised similarly, avoiding excessive deficit or deficiency.

2. Soil analyses

Soil samples were taken from each hole that was used for studying root growth and distribution. Soil was collected from four depths, i.e., 0–30 cm, 30–60 cm, 60–90 cm, and 90–120 cm. Physical and chemical analyses of the soil were done by two commercial laboratories (LabServe Micro,

Stellenbosch, and LabServe Analytical Services, Nelspruit, South Africa).

3. Root growth and starch analyses

Root systems of the treatments were studied during spring using the well-known root profile method of Böhm (1979), whereby a trench of 2.0 m deep was dug parallel to the vine row and 300 mm from the vine trunk. Three replicates per treatment were used for root studies, and two holes were made per treatment replicate (to a total of 90 holes). After careful exposure of roots, a grid system of 200 × 200 mm was set up against the profile wall. Roots inside the grid squares were plotted vertically to a depth of 1.8 m and laterally to the treatment space boundaries of adjacent vines. To increase accuracy, roots of the very narrow spacing treatments were plotted across the vine root system boundaries (three vines for 0.3 m spacing and two vines for 0.6 m spacing). Roots were plotted in five root diameter categories, i.e., fine (<0.5 mm), extension (0.5–2 mm), permanent (2–5 mm), and framework (5–10 mm, and >10 mm) roots (Richards, 1983). To simplify data, numbers within classes were combined, and the classes were reduced to three, namely <0.5–2 mm, 2–5 mm, and >5 mm. The starch content of <0.5–5 mm thick roots was determined enzymatically in freeze-dried material (Hunter *et al*., 1995a), sampled just after completion of the root study.

4. Physiological measurements

Physiological measurements were done three times during the growth season at random in the centre of two of the four block replications, i.e., pre-véraison at approximately pea berry size and post-véraison at approximately six weeks and two weeks before harvest, respectively.

Ambient and interior-canopy photosynthetic active radiation (PAR) (400–700 nm, µmol/m^2/s) was captured in the bunch zone and upper third of the canopy using an LI-COR LI-191 Line Quantum sensor during mid-morning and afternoon, starting at 10:30 and 14:00 and expressed as % of ambient; this was repeated in three different canopies per replication. Photosynthetic activity (and related measurements) and stem water potential were measured on two basal primary shoot leaves in the bunch zone per replication. Photosynthetic activity (µmol CO_2/m^2/s), relative humidity (%), air temperature (°C), leaf temperature (°C), photosynthetic photon flux density (PPFD) (µmole/m^2/s), transpiration (mmole/m^2/s), stomatal conductance (mmole/m^2/s), and internal [CO_2] measurements started at 10:30 using an open system LI-COR LI-6400 portable photosynthesis meter at gas flow of 400 ppm CO_2 and equipped with a LI-COR 6400-02B red/blue light source adjusted to 2000 µmol/m^2/s. Stem water potential (-kPa) measurements were done over mid-day, using two equally calibrated (flow rate of 1000 kPa/30 s) Scholander pressure chambers (Scholander *et al*., 1965); leaves were covered for at least one hour before the start of measurements. Diurnal–Nocturnal cycles (24 hours) of leaf temperature, leaf photosynthetic and dark respiration activity, and stem water potential (as described above) were done on one replication only during the post-véraison period at approximately six weeks and two weeks before harvest, respectively, measuring every four hours (from 09:00–09:00) on two basal, bunch zone located leaves per replication. Average values of the two measuring times and three years of measurement are presented.

Leaf chlorophyll content was measured pre-véraison and post-véraison in three random parts of the canopies of two of the four replications on leaves on the outer/exterior sides of the canopy and in the middle/interior part of the canopy at 10:30 and 14:00. One leaf positioned on the front (exterior) side and one leaf in the middle (interior) of the canopy were measured. All three measurements per canopy part were done at the same time. Leaf chlorophyll content was measured with an Opti-sciences CCM-200 clip-on device.

Berry pulp temperature was measured pre-véraison and post-véraison in three random parts of the canopies of two of the four replications on bunches carried on the outer/exterior sides of the canopy and in the middle/interior part of the canopy at 10:30 and 14:00. For the outer hanging bunches, one berry positioned on the front (exterior canopy) side and one berry on the back (interior canopy) side of the bunch were measured. One berry per interior canopy bunch was also measured. Berry temperature point measurements were done using an ETI 2202 hand-held thermometer (Electronic Temperature Instruments Ltd, Easting Close, Worthing, West Sussex, UK) fitted with a probe.

5. Statistical design and methods

The experiment was laid out as a randomised block design with 15 spacing treatments and four replications. The Shapiro–Wilk test verified the normality of the standardised residuals from the model (Shapiro and Wilk, 1965). Levene's test confirmed homogeneous standard deviations (Levene, 1960). Data were subjected to ANOVA using the General Linear Models Procedure (PROC GLM) of SAS software (Version 9.4; SAS Institute Inc, Cary, USA) to determine polynomial trends. Where applicable, data was then either subjected to the regression procedure (PROC REG) to fit linear and quadratic regressions or to the non-linear regression procedure (PROC NLIN). A join point regression was fitted to the data to describe the changes in trends over vine spacing. Join point regression, also known as change point regression, broken stick, or segmented regression, assumes that data can be divided into subsets—each with their own unique linear trend (Hudson, 1966). PROC NLIN was performed to fit more than one line to the data to determine the "join point/s". Outliers were determined using the Shapiro-Wilk test on standardised residuals from the model to verify normality (Shapiro and Wilk, 1965). Outliers were replaced by predicted values of the model. Fisher's least significant difference was calculated at a 5 % level to compare the means of the factors (main effects) and factor interaction means (Ott and Longnecker, 2010). A probability level of 5 % was considered significant for all tests.

TABLE 1. Soil physical and chemical characteristics of the loam soil in the Breede River Valley, Robertson, in which the Shiraz/101-14 Mgt vineyard was planted to different vine spacing.

Spacing (m)	Bulk density (kg/m³)	Stone v/v (%)	Clay (%)	Silt (%)	Sand (%)	Sand Fine (%)	Sand Medium (%)	Sand Coarse (%)	WHC (mm/m)
0.3	1509.0	7.05	14.62	9.07	76.32	35.33	14.12	26.86	99.95
0.6	1518.7	6.33	17.13	8.46	74.42	38.19	14.40	21.83	103.41
0.9	1503.1	12.35	15.38	8.33	76.29	27.98	16.41	31.90	83.25
1.2	1508.4	7.62	14.13	9.08	76.79	36.87	14.86	25.07	100.72
1.5	1558.9	6.15	18.67	8.58	72.75	33.58	14.43	24.74	97.93
1.8	1558.5	7.86	16.23	9.16	74.61	30.64	16.41	27.56	91.76
2.1	1514.3	12.24	14.64	8.49	76.87	32.53	15.44	28.90	89.52
2.4	1527.2	7.76	11.29	9.29	79.42	43.28	13.16	22.97	109.92
2.7	1531.6	5.46	20.54	8.87	70.59	36.08	14.32	20.20	101.87
3.0	1537.8	8.74	13.83	9.33	76.83	39.74	13.16	23.93	104.48
3.3	1518.7	6.67	14.47	7.98	77.56	33.14	15.63	28.79	95.57
3.6	1499.3	10.27	13.66	9.63	76.72	35.64	15.11	25.97	96.61
3.9	1515.3	5.52	14.08	9.25	76.67	38.85	14.37	23.45	105.94
4.2	1538.4	7.98	15.08	9.55	75.38	36.00	16.39	22.99	98.59
4.5	1554.6	5.17	16.85	7.87	75.28	39.97	13.97	21.34	106.78

Spacing (m)	Base saturation (%)				Exchangeable cations (cmol/kg)				T-Value (mg/kg)
	Na	K	Ca	Mg	Na	K	Ca	Mg	
0.3	5.61	2.16	48.56	20.60	0.84	0.33	12.84	3.63	28.55
0.6	7.72	2.72	46.00	21.02	0.90	0.34	10.12	3.59	30.04
0.9	7.83	3.06	46.96	21.92	1.16	0.44	9.61	4.65	28.23
1.2	5.12	1.88	48.88	20.70	0.97	0.37	14.70	4.55	29.01
1.5	9.38	2.92	45.39	21.16	1.09	0.36	8.38	3.47	29.77
1.8	7.14	2.23	48.92	19.89	0.85	0.29	10.70	3.14	28.46
2.1	7.75	2.50	47.58	21.43	1.19	0.41	12.26	4.74	28.69
2.4	6.06	1.66	47.38	20.12	1.06	0.28	14.24	3.82	30.77
2.7	7.37	2.15	47.60	20.35	0.97	0.26	10.99	3.32	29.42
3.0	6.08	1.69	48.74	20.71	1.00	0.27	12.63	3.85	28.54
3.3	11.20	3.18	43.37	22.05	1.13	0.33	5.79	3.40	29.40
3.6	7.78	2.32	48.73	19.77	1.03	0.30	10.86	3.40	28.73
3.9	6.92	2.33	48.06	21.15	0.83	0.30	10.67	3.55	28.14
4.2	7.65	2.82	45.73	21.15	0.91	0.37	10.26	4.24	30.18
4.5	11.25	3.04	50.08	22.64	1.01	0.28	6.53	2.68	23.60

Spacing (m)	Resistance (ohm)	pH (KCl)	C (%)	P (Ambic I (mg/kg)	S (mg/kg)	Na (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)
0.3	598.90	6.68	0.27	13.20	15.47	192.50	127.8	2567.1	435.4
0.6	592.82	6.97	0.28	14.91	34.52	207.75	133.5	2023.7	431.3
0.9	524.70	6.89	0.24	17.08	34.59	265.95	173.5	1922.7	558.4
1.2	609.28	7.32	0.15	14.28	13.38	223.04	143.3	2940.9	545.4
1.5	523.55	6.83	0.18	13.41	45.11	251.55	141.5	1676.8	416.9
1.8	522.01	6.95	0.27	17.95	23.43	196.41	113.2	2139.4	376.7
2.1	497.66	7.02	0.26	13.82	37.61	274.14	160.9	2452.1	569.1
2.4	606.07	7.30	0.24	13.80	19.79	244.33	109.0	2847.6	458.2
2.7	647.13	7.28	0.16	14.89	28.44	223.30	103.2	2198.2	397.9
3.0	525.93	7.22	0.24	15.93	37.59	231.11	104.8	2525.1	461.6
3.3	512.11	5.70	0.24	11.98	41.33	259.21	127.8	1157.7	407.5
3.6	597.04	7.01	0.17	12.52	31.73	237.34	118.0	2171.3	408.5
3.9	562.81	7.02	0.29	13.34	14.17	190.08	117.5	2134.7	425.8
4.2	606.63	6.93	0.23	10.23	16.81	209.46	144.1	2052.1	508.9
4.5	569.24	6.54	0.16	11.63	41.86	231.64	108.3	1305.5	321.3

Values represent the whole 0–120 cm soil profile (average of separate 0–30 cm, 30–60 cm, 60–90 cm, and 90–120 cm analyses); WHC = Water Holding Capacity; T-Value = Total exchangeable cations plus H^+.

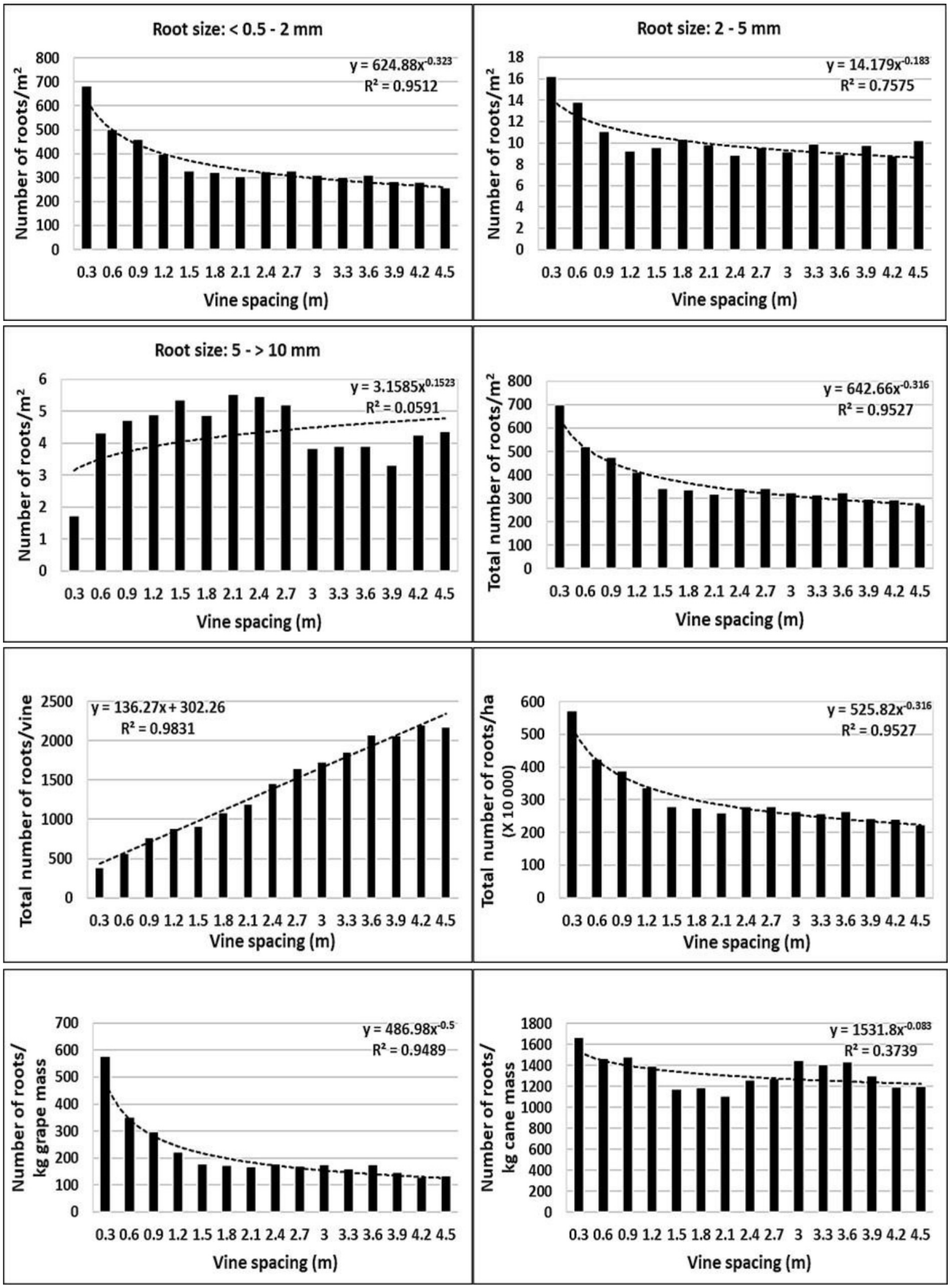


FIGURE 1. Vine spacing effect on the development of different root sizes, the total number of roots, and the ratios between roots and grape and cane mass of Shiraz/101-14 Mgt.

RESULTS AND DISCUSSION

1. Soil conditions

Average values of basic physical and chemical soil parameters in the 0–120 cm soil profile (average of separate 0–30 cm, 30–60 cm, 60–90 cm, and 90–120 cm analyses) are presented in Table 1. The generally neutral pH soil was classified as loam soil with relatively good resistance. No significant numbers of soil-borne pests that may have affected vine health were found (data not shown). The bulk density of the soil was remarkably uniform with depth (data not shown). It was similar across treatments and was generally close to the 1.5 g/cm^3 norm, beyond which root penetration declines (Richards, 1983). In general, the soil analyses showed that vines of the different treatments were exposed to relatively uniform soil conditions that would have imposed little if any, irregular additional effects on treatment outcomes.

2. Root development

The study was executed in a terroir characterised by moderately fertile soil and a relatively hot summer climate

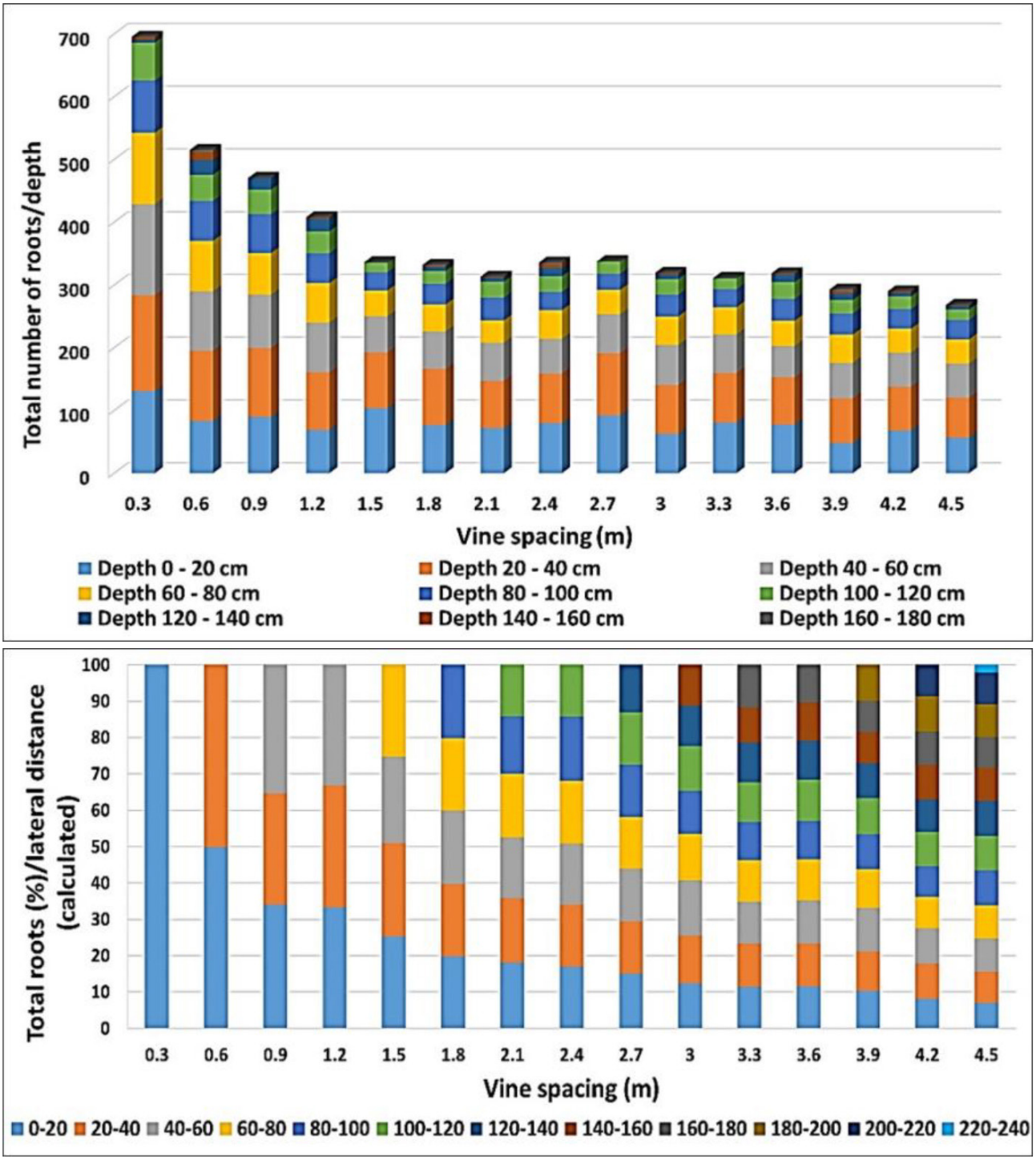


FIGURE 2. Vine spacing effect on the development of roots in different soil layers (vertically and laterally) for Shiraz/101-14 Mgt.

with low winter rainfall. The rootstock used is of moderate vigour (Southey, 1992), vines were not overly irrigated and fertilised, and canopies were well accommodated. Root development of the different vine spacing treatments over an 11-year period of the experiment is shown in Figure 1. An increase in root density/m^2 soil profile was found with closer spacing, particularly in the fine, extension and permanent categories, thereby increasing the total number of roots/m^2 and confirming trends found with previous research under different climatic, soil and cultivation conditions (Archer and Strauss, 1985; Hunter, 1998a; Archer and Hunter, 2004; Hunter *et al*., 2016a). Irrespective of treatment, the soil profile was more than 90 % dominated by roots with diameters of less than 2.0 mm, showing the importance of fine roots in effective soil exploitation. This was also found by Archer *et al*. (1988) for different trellising systems, regardless of the size of the root system. The total number of roots per soil profile increased linearly with wider spacing on a per-vine basis, confirming the positive relationship between aboveground and subterranean growth (Archer and Strauss, 1985; Archer *et al*., 1988; Hunter, 1998a; Archer and Hunter, 2004). It followed a decreasing trend per ha, showing a much higher soil root population for narrowly spaced (<1.5 m) vines. Considering that this general trend has also been observed under very different growth conditions (Champagnol, 1979; Richards, 1983; Archer and Strauss, 1985; Hunter, 1998a; Archer and Hunter, 2004), this would be beneficial for vines planted in poor/infertile soils and under unfavourable climatic conditions, e.g., in high temperature, low rainfall conditions, to increase the explorative and absorptive capacity of the root system and to support shoot growth. Colonisation of available soil volume by the roots is critical to dictate aboveground:belowground growth balances. Notwithstanding very low numbers of roots in the category, the larger framework roots in the soil profile decreased noticeably beyond 2.7 m spacing. This may point to a shift in energy/carbon balance in the vines to maintain the generation of new roots instead of the build-up of reserves, to increase water and nutrient absorption to supply the demand from aboveground biomass (vegetative and reproductive growth), as well as to avoid stress inflicting soil and canopy conditions. Considering the number of roots in the soil profile per grape mass and cane mass (data on grape and cane mass not included in this paper), the optimum appeared to be reached at approximately 1.8 m spacing, followed by a plateau, and further change with very wide spacing. The number of roots/kg grape mass varied from 600–100 (and 1600–1100/kg cane mass, this being more stable according to vine size/spacing) from the narrowest to the widest spacing, with approximately 180 occurring for a wide range from 1.5–3.6 m vine spacing and that for 1.2 m spacing and narrower being more than 200. The pattern across spacing treatments of the number of roots/kg grape mass showed a remarkable correspondence with the number of fine roots/m^2 soil profile. Although these numbers are not universally applicable (because of likely effects of soil type, water, nutrition, variety, rootstock, climate, etc., on general growth and root system dynamics), they should be judged as indicators of what can be expected under similar growth conditions; the importance of a proper root system to support shoot growth and grape development; and the necessity of creating judicious aboveground:subterranean growth ratios/balances along with crop load (Hunter *et al*., 2016a).

In general, the highest concentration of roots seemed to occur in the 0–60/80 cm deep soil profile (Figure 2), confirming preferential rooting zone results from other studies under different cultivation conditions (Van Zyl and Weber, 1981; Champagnol, 1979; Swanepoel and Southey, 1989; Hunter and Le Roux, 1992). Beyond a depth of 120 cm, the number of roots decreased markedly. Narrower spaced vines (<1.5 m) showed higher root numbers in deeper soil layers, demonstrating that their roots indeed penetrated deeper soil layers (most likely at a steeper angle), thereby increasing the preferential root zone, as also found previously (Archer and Strauss, 1985; Hunter, 1998a). Interestingly, despite the root density differences amongst the spacing treatments, the lateral distribution of roots was almost uniform for each spacing within their separate confined spaces. Root density and depth appeared to be most affected by spacing. The results confirm the steering effect of vine spacing on root system development. It validates the hypothesis that deeper and more fertile loamy soil conditions in this experiment, together with lateral space restriction induced by the root systems of adjacent vines, allowed and forced the roots to compensate and explore deeper soil layers, especially in the case of narrower-spaced vines. The data affirms the dynamic, responsive nature and regenerative ability of the grapevine root system (Richards, 1983; Hunter and Le Roux, 1992; Southey, 1992; Hunter *et al*., 2016a).

In previous comprehensive vine spacing research done on Pinot noir/99 Richter (Archer and Strauss, 1985; Hunter, 1998a), both vine spacing and row spacing were changed, whereas in this experiment on Shiraz/101-14 Mgt, vine spacing was the only variable. Other major differences between the previous and this research (except for variety and rootstock) lie in soil fertility and depth as well as irrigation volume. The earlier studies were done on a medium potential, relatively shallow soil, and vines were either being grown under dryland conditions (Archer and Strauss, 1985) or only being supplementarily irrigated at pea size and véraison (Hunter, 1998a). Like in this research, these studies also showed that the lateral limitation (because of the competitive presence of the root system of adjacent vines) of closely spaced vines on soil volume availability and utilisation steered the root system towards a more vertical penetration angle to explore deeper soil layers. Wider spacing resulted in more horizontal development, and intra-row space also had a higher restrictive bearing on root system distribution than inter-row space despite the soil type differences between the studies. The closer the vines were spaced, the more the density of the root system and ratio of fine:thick roots increased; this also led to naturally higher water and nutrient absorption by the root system, which may also encourage

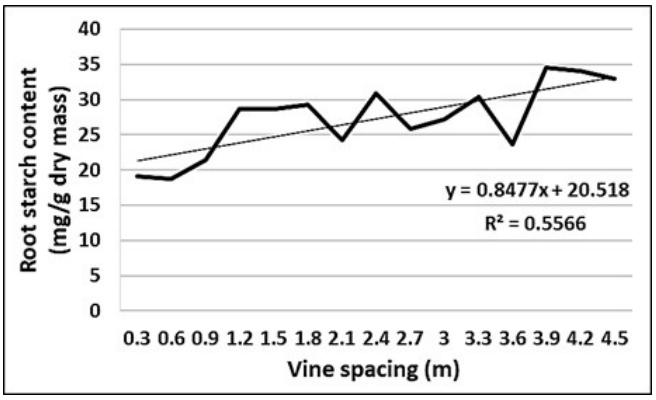


FIGURE 3. Vine spacing effect on the starch content of <0.5–5 mm thick roots of Shiraz/101-14 Mgt.

deeper root penetration towards the higher water contents of deeper soil layers. As in the previous study (Hunter, 1998a), a higher vine starch depletion or lower starch accumulation was found in this study with closer spacing (Figure 3). A gently increasing starch content occurred with wider spacing.

In addition to being favourable to water and nutrient absorption, a root system with a higher fine root component increases hormone production and availability, such as cytokinin, gibberellin, and abscisic acid, thereby regulating shoot and fruit development and photosynthetic activity and synergistically or antagonistically affecting availability and action of other hormones, e.g., auxin (Freeman, 1983; Richards, 1983; Davies *et al*., 1986; Field *et al*., 2009). A soil environment-induced root stress enhancement (e.g., via depletion of water and nutrients or other soil-environment-related stresses) may stimulate abscisic acid transport in the xylem sap but may impair cytokinin formation and transport, thereby governing many aspects of shoot physiology, like stomatal movement, transpiration, photosynthetic activity, and whole plant water relations, including possible embolism repair coupled to aquaporin activation/formation, amongst others (Davies *et al*., 1986; Hunter, 1998b; Stoll *et al*., 2000; Lovisolo *et al*., 2010; Gambetta *et al*., 2020). Under high irradiance, vapour pressure deficit and evapotranspiration conditions, this may help to maintain hydraulic integrity and avoid cavitation in the xylem. This may also help the variety (Shiraz) to sustain so-called isohydric behaviour [questioned by Hochberg *et al*. (2018), based on multiple biotic and abiotic impact factors on grapevine physiological behaviour], despite its general description as being anisohydric and the rootstock (101-14 Mgt) being characterised as drought-sensitive (Southey, 1992). Recognising the complexity of the grapevine and the magnitude of different impact factors on growth and development, Lovisolo *et al*. (2010) listed evidence of varieties behaving isohydrically and anisohydrically to avoid stress and to increase the efficiency of water use. It would be more plausible to describe varietal behaviour within the context of prevailing environmental conditions and physiological status, as well as the ability to survive and recover from stressful conditions, rather than group them categorically.

It follows that a more active canopy would stimulate the synthesis and transport of auxin to roots, adding to the root-derived auxin pool that orchestrates growth and gravity responses in this department (Overvoorde *et al*., 2010). Cytokinin is considered a potential anti-senescence hormone that regulates the growth and cell division of plants (Rivero *et al*., 2007). It showed rapid decline under drought stress conditions, changing growth and development (Yang *et al*., 2002; Kudoyarova *et al*., 2019). An active regenerative root system would thus increase cytokinin production and water absorption capabilities. This may be beneficial under water deficit conditions to increase drought tolerance as an effective, natural mechanism, buffering turgor loss, the physiological stress stimulated formation of abscisic acid, and suppressing or delaying drought-induced leaf senescence, with continuation of functional photosynthetic activity and water use efficiency (Hunter, 1998b; Rivero *et al*., 2007). In addition, an active root system is required to supply potassium, an essential partner to abscisic acid in stomatal control and physiological homeostasis (Patrick, 1997; Carlomagno *et al*., 2018; Gambetta *et al*., 2020). However, under water deficit conditions, sucrose phloem loading should also be sustained by active leaf photosynthetic function to prevent the accumulation of excessive potassium in the phloem, which may be loaded to maintain phloem turgor when sucrose formation is insufficient (Giaquinta, 1983; Patrick 1997; Hunter and Ruffner, 2001). From a grape quality perspective, an abundance of potassium in grapes is generally not preferred and may reduce acidity and increase pH (Hunter and Ruffner, 2001).

Water stress had been shown to decrease the apoplastic water fraction, specific leaf weight, starch levels, and soluble carbohydrate concentration (Rodrigues *et al*., 1993; Patakas and Noitsakis, 1999), while water-stressed shoots showed lower hydraulic conductance per unit length and lower specific conductivity in internodal and foliar xylem (Schultz and Matthews, 1993; Lovisolo and Schubert, 1998). The leaf osmotic adjustments enabled the maintenance of turgor but still reduced carbon assimilation. According to Escalona *et al*. (2012), soil moisture depletion in the root zone led to lower root respiration rates, which may be negative for root metabolic activity but may lead to a lower portion of photosynthetically derived carbon being lost. Both canopy and root activity should, therefore, be maintained to secure an osmotic gradient for continued water and carbohydrate transport and whole-plant metabolic activity. Plant strategies to avoid or tolerate drought stress, in other words to reduce water deficit vulnerability and protect hydraulic integrity, include stomatal closure, sink/source adjustment, such as increasing root growth and decreasing canopy growth, accelerated leaf senescence and abscission of older leaves, and avoiding embolism formation in a segmented way in at least the permanent structure (Fischer and Turner, 1978; Levitt, 1980; Zimmermann, 1983; Epstein and Bloom, 2005; Pou *et al*., 2012; Hochberg *et al*., 2017). Furthermore, in

grapevines, other mechanisms, such as suberin accumulation in response to water stress were observed in roots (Vandeleur *et al*., 2009) and around vascular bundles in petioles (Hochberg *et al*., 2013), serving as apoplastic barriers to water loss, with low expression of aquaporin isogenes, decreasing hydraulic conductance across the root surfaces, and thereby modulating stress responses (Gambetta *et al*., 2013). Exposure to environmental conditions at the vineyard level over many years (e.g., in the case of old vines) would allow morphological and physiological adaptations and greater plasticity of the individual vines to mitigate the (sudden) impact of factors of extreme nature. Under such a magnitude of variables changing from vineyard to vineyard, it is hardly possible to demarcate the value ranges of physiological parameters that are so often listed in the literature.

An increase in root density may lead to higher localised water depletion per soil volume, which may be enhanced in environments/seasons with very low rainfall and/or where high ambient summer temperatures prevail (the latter as per location of this experiment—Hunter *et al*., 2016b). Although a lower leaf water potential would be expected for narrow spacing in such conditions, compared to wider spacing, vines may react by closing stomata to reduce water loss *via* transpiration under water deficit conditions (Archer and Strauss, 1989; Lovisolo *et al*., 2010). However, under moderate water supply, growth may be stimulated, which may lead to higher total water loss, but also a more shaded canopy that, in turn, would have a further restrictive effect on leaf activity (Kriedemann, 1968; Düring, 1991; Hunter *et al*., 2014; Hunter *et al*., 2020) when vines are very closely spaced. On the other hand, a more open canopy would naturally lead to higher leaf activity unless it is too open, light energy is lost, leaf senescence and photo-inhibition occur, water stress prevails, and stomatal movement is restricted (Hunter and Visser, 1989).

Root system growth adaptation and physiological control over aboveground growth play an important role in the survival ability of the scion variety but also impact heavily on sustainability (Archer and Hunter, 2004). Source:sink relationships play an integral role in plant response and may push the boundaries of normal stress avoidance mechanisms of leaves and roots when demand for carbohydrates (e.g., in the form of higher crop load) is high. A well-expanded root system would always increase the ability of the vine to rapidly recover water status and carbon gain after hydraulic stress. Medrano *et al*. (2015) found that respiratory losses by roots, leaves and grapes of potted Tempranillo and Grenache were linked to water status, increasing with stressful conditions. The complexity/architecture and composition of the canopy, including leaf position and age as well as irradiance, regulating photosynthesis and transpiration, has a major role in the physiological control of whole plant carbon balances (Medrano *et al*., 2015; Hunter *et al*., 2020). Stress avoidance and improved water use efficiency by means of better choices and adaptation of scion and rootstock, site selection, and cultivation practices are clearly very important factors in the governing of future climate scenarios for grape production and quality (Hunter *et al*., 2016a). Grapes of many varieties are prone to significant transpirational water loss during late ripening (Greer and Rogiers, 2009; Carlomagno *et al*., 2018) and the aforementioned factors should, thus, have high priority in cultivation.

3. Microclimate and Physiological parameters

The diurnal–nocturnal cycles of leaf temperature, stem water potential, and leaf CO_2 gas exchange of the different plant spacing treatments are shown in Figure 4. Leaf temperatures of the treatments increased during the day from 09:00 until 17:00, where after it decreased sharply to the lowest values at 05:00. Stem water potential followed in tandem with values decreasing towards late afternoon, after which the vines recovered to highest values occurring early morning, like that found by Naor and Wample (1996). The leaf CO_2 gas exchange followed a similar trend to stem water potential, decreasing towards late afternoon and into the night, reaching most negative values (representing dark respiration) before midnight, with values of around zero after that until 05:00. Respiration rates seemed affected by the diurnal build-up of photosynthetically generated carbohydrate (Escalona *et al*., 2012). The results agree with those found by Zufferey (2016). At this stage of the season, during the grape ripening period, basal leaves that were measured displayed low photosynthetic activity, albeit with assimilatory competence that would sustain maintenance metabolism, while increasing the hoarding of sucrose instead of translocation (Hunter *et al*., 1994; Hunter *et al*., 2016b).

Light microclimate (photosynthetic active radiation — PAR) in the upper and lower zones of the canopy followed an increasing trend from narrow to wide spacing (Figure 5), indicating a difference in canopy density between narrower and wider spaced vines, regardless of the uniform spur distribution across treatments. Mid-day stem water potential values (negative) at pre- and especially post-véraison stages followed an ostensible decreasing trend, i.e., increasing water stress, towards wider vine spacing (Figure 6). Parallel to this and despite the general increase in PAR (Figure 5) and leaf chlorophyll index (during grape ripening) (Figure 7) with wider spacing, photosynthetic activity showed very little movement with a change in spacing (Figure 8). As also demonstrated in previous work (Hunter and Visser, 1989), leaf chlorophyll content increased with better canopy microclimate conditions during post-véraison, from narrow to wide spacing; it was, however, found not to be a reliable indicator of photosynthetic activity. The latter seemed rather driven by source:sink relationships and other competitive interactions amongst leaves (Hunter, 2000). Considering the PAR and stem water potential trends, the photosynthetic activity of the leaves seemed to display the respective contrasting (and, in this case, counteracting) effects of PAR and water potential, the former generally having stimulating and the latter having limiting effects on photosynthetic activity. Although an ageing canopy towards harvest time would have been a secondary contributing element to both effects, these two generally opposite (Poni *et al*., 1994;

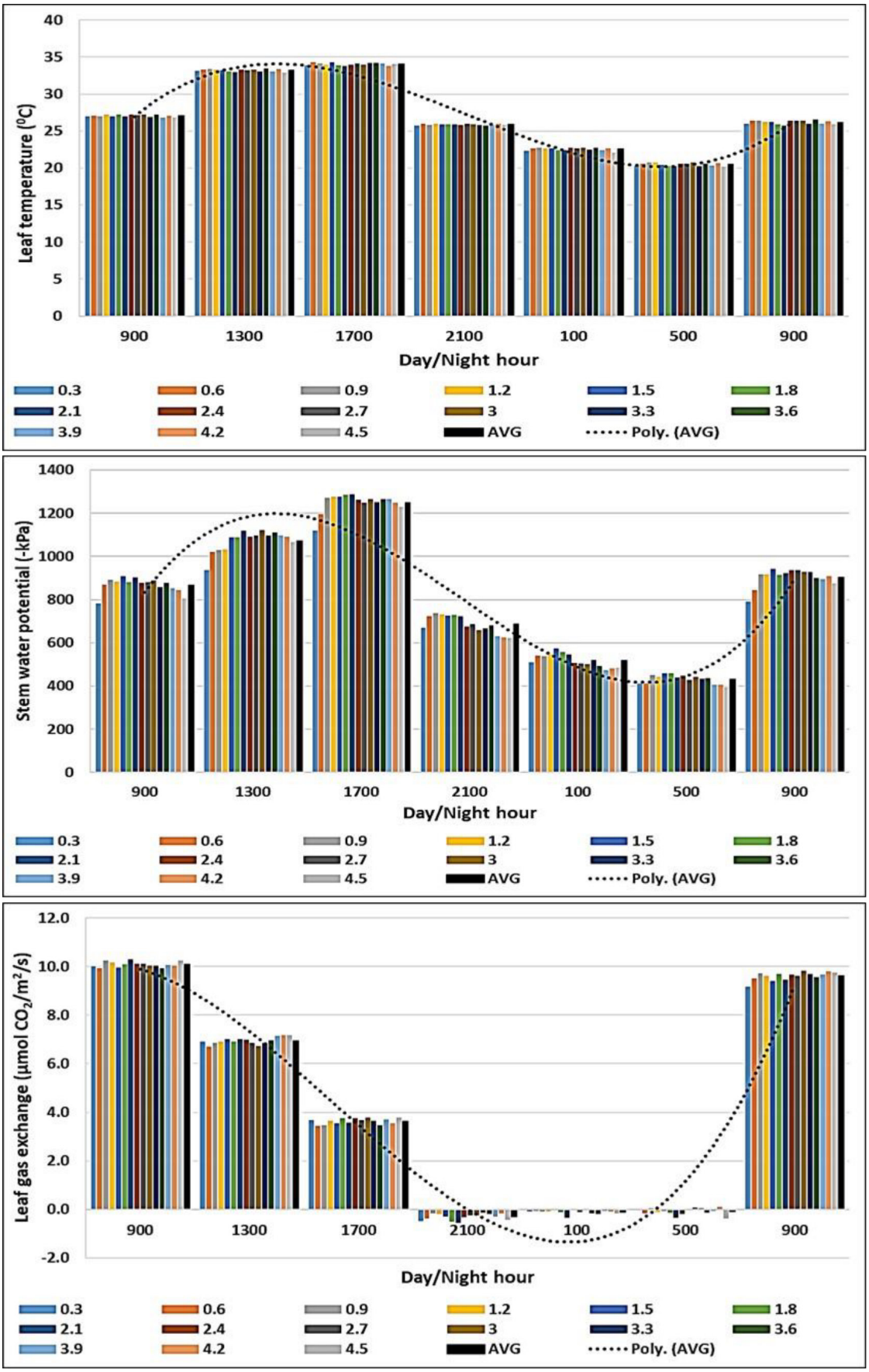


FIGURE 4. Average diurnal–nocturnal cycles of leaf temperature, stem water potential, and leaf CO_2 gas exchange measured during the grape ripening period of Shiraz/101-14 Mgt (means of 2018–2020 seasons).

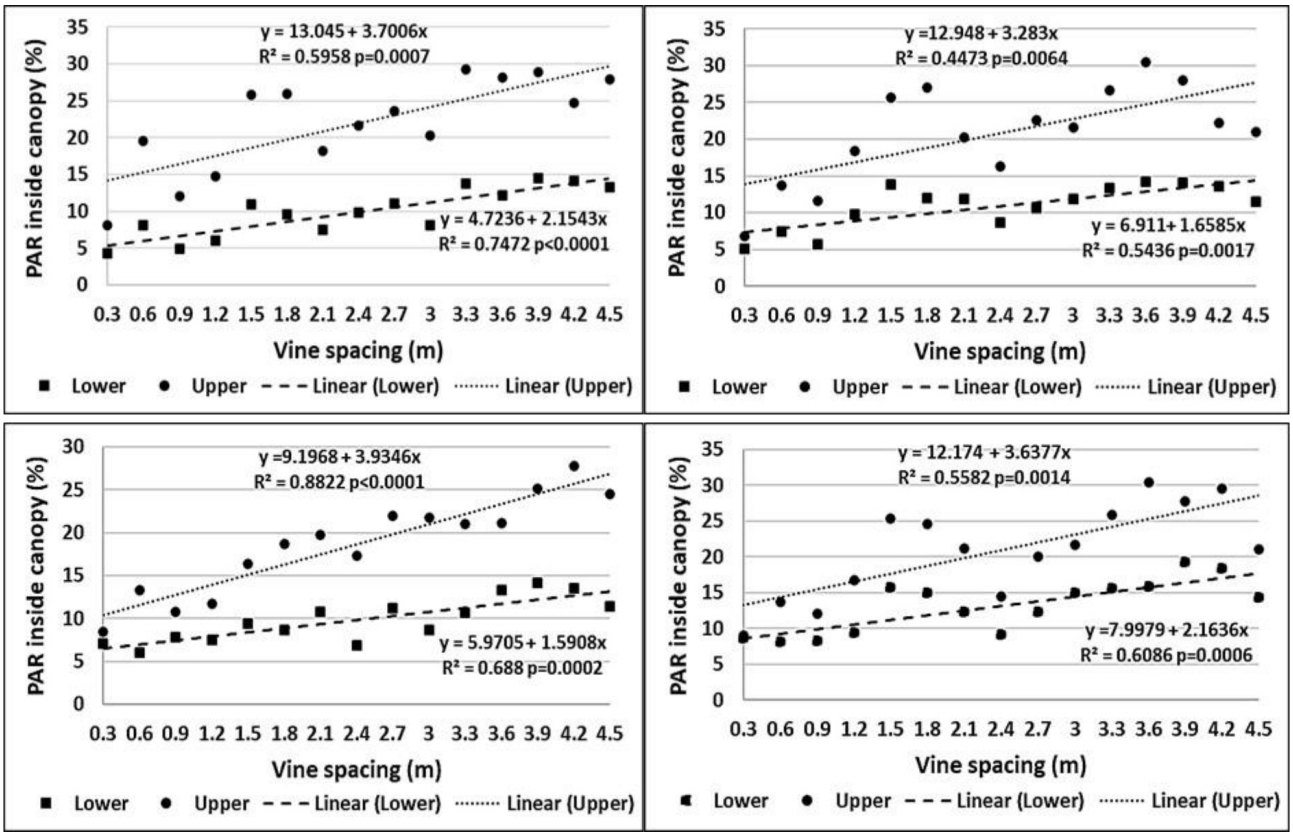


FIGURE 5. Photosynthetic active radiation (PAR) (% of ambient – measured as µmol/m2 /s) [pre-véraison (top) and post-véraison (bottom); morning (left) and afternoon (right)] in the bunch zone and upper zone of the canopy of Shiraz/101-14 Mgt (means of 2014–2020 seasons).

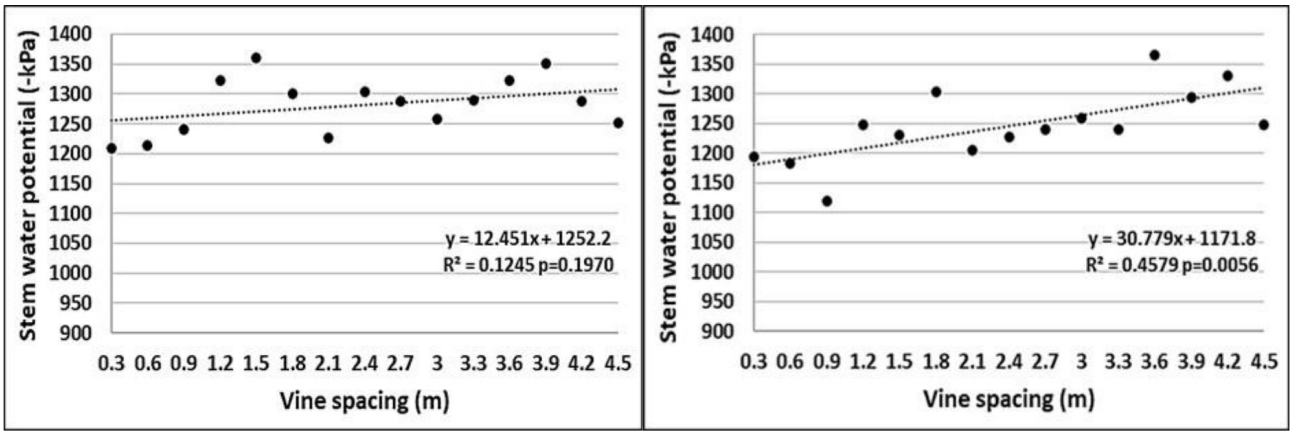


FIGURE 6. Stem water potential [(pre-véraison (left) and post-véraison (right)] of Shiraz/101-14 Mgt (means of 2014–2020 seasons).

Hunter *et al*., 2014) impact factors apparently had an almost equalising consequence for the photosynthetic activity of the leaves.

Stomatal conductance (slightly decreasing), transpiration (slightly increasing) and vapour pressure deficit (slightly increasing) trends with wider spacing (data not shown) paralleled the findings on photosynthetic activity during pre- and post-véraison periods. At the same time, the internal [CO_2] decreased with wider spacing, whereas leaf temperature in the bunch zone fluctuated between 31 °C and 37 °C, with an increasing trend from narrow to wide treatments (data not shown). The predicament of the vines surfaced from the amount of water that transpired per photosynthetic output. Despite the better-exposed canopies of wider-spaced vines, water use efficiency showed a straight, horizontal line from narrow to wide spacing during both the active growth period and later stages of grape ripening when canopies were already senescing (data not shown). These trends indicate

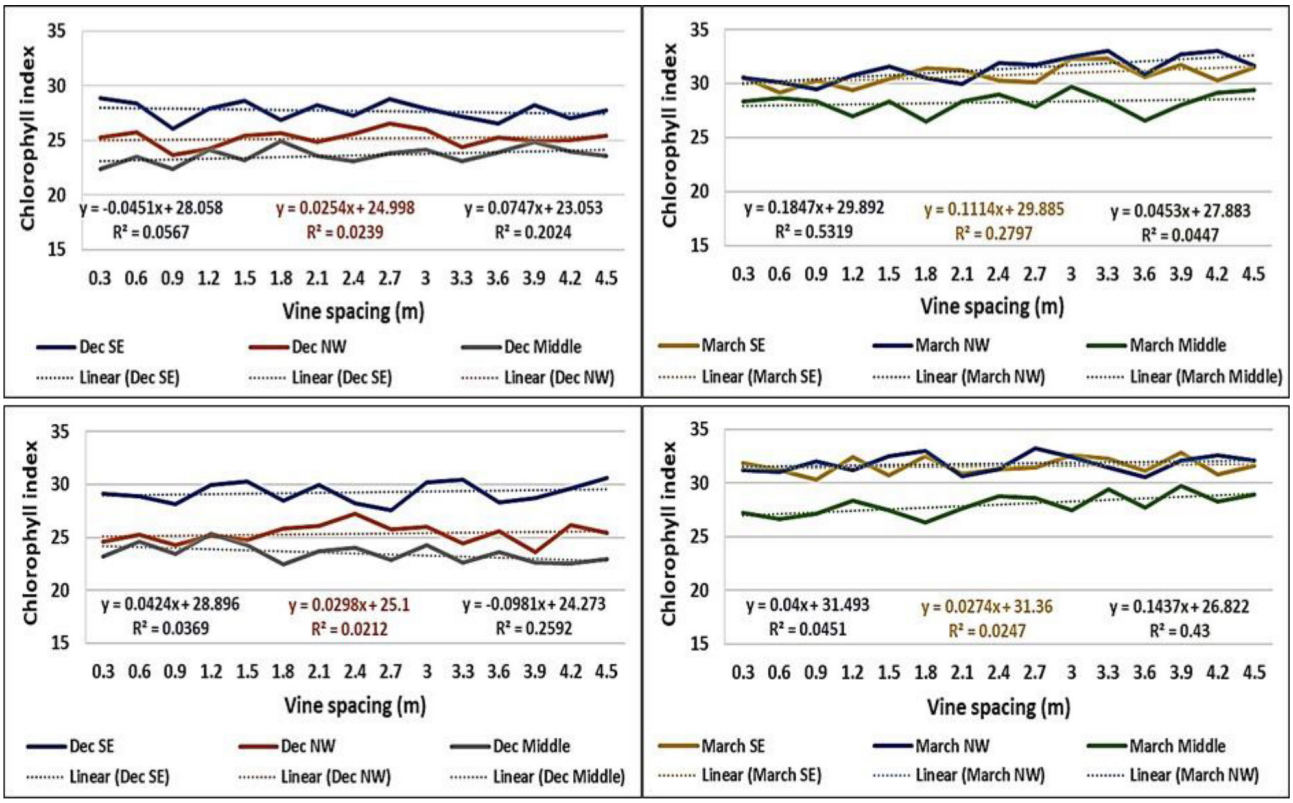


FIGURE 7. Leaf chlorophyll index measured in the morning (top) and afternoon (bottom) on leaves on the exterior/front of different canopy sides and in the interior/middle of the canopy [(pre-véraison (left—December) and post-véraison (right—March)] of NNE–SSW oriented Shiraz/101-14 Mgt (means of 2017–2020 seasons).

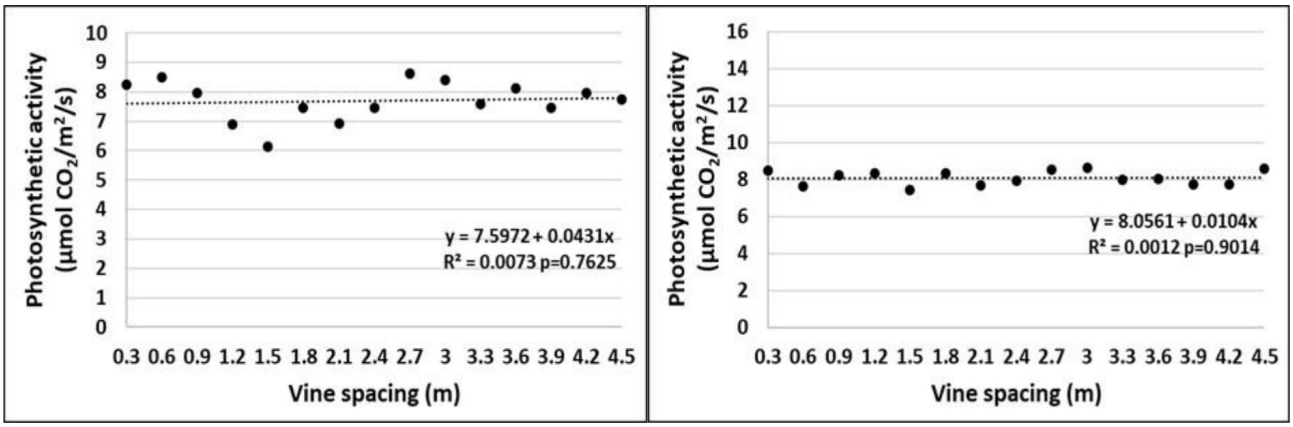


FIGURE 8. Photosynthetic activity [(pre-véraison (left) and post-véraison (right)] of Shiraz/101-14 Mgt (means of 2014–2020 seasons).

physiological compensatory reactions that likely restricted the utilisation of available water for photosynthetic activity with wider spacing despite the (ostensible) positive driving force for water transport. The decrease in internal [CO_2] and increase in leaf temperature with wider spacing points to strict stomatal control over gas exchange and transpirational water loss, to the apparent detriment of photosynthetic activity and leaf cooling. A close correlation between photosynthetic activity and stomatal control is known to exist under challenging environmental conditions and, depending on the phenological stage, also affects recovery after the release of water stress (Flexas *et al*., 2002). The increase in leaf temperature may also indicate failure to respond to and guard against excessive direct radiation onto leaves, i.e., a more diaheliotropic (perpendicular to the sun) orientation was maintained, whereas paraheliotropic movement that orientates the leaf parallel to the direction of the sun to avoid excessive radiation, may have been restricted (Long *et al.*, 1994); the latter might have become more and more difficult with the loss in elasticity of leaf petioles and laminae as the canopy aged. This also means, as found with *Vitis californica* (Gamon and Pearcy, 1989), that excessive leaf temperature

(together with a low stem water potential) might have become an increasingly important factor driving senescence and limiting carbon gain and distribution as the growth season progressed. In a study involving four row orientations in the same location as this study, it was shown that leaf lamina orientation is a reality in a grapevine canopy, demonstrating drastic changes from just after véraison to just before harvest for different vineyard row orientations (Pisciotta *et al*., 2011). Similar responses were found in other plants (Fischer and Turner, 1978; Werner *et al.*, 1999). A physical lack of space, interior canopy shade, photo-morphogenesis, phytochrome effects and a demand for increased photosynthetic output may be reasons for this. Feng *et al*. (2002) argued that high irradiance and low water availability for transpiration may lead to insufficient thermal dissipation in a leaf to protect physiological processes (including the photosynthetic apparatus) from thermal- and photo-destruction, leading to photochemical inefficiency. A photoprotective role of photorespiration could be assumed under such conditions when accompanied by high temperatures, responding to low intercellular [CO_2] and stomatal conductance and deficiencies in NADPH and ATP supply (Guan *et al*., 2004; Lovisolo *et al*., 2010). As during mobilisation of reserve carbohydrate under reduced source:sink ratios (Candolfi-Vasconcelos *et al*., 1994; Hunter *et al*., 1995b), anaplerotic processes such as vacuolar-storage release and gluconeogenesis may also be involved, as a constant supply of carbohydrate would still be demanded to maintain osmo-regulation and to drive the activity of the root system, the leaves, and the grape development and ripening process under stressful, carbon-starved conditions (Hunter, 2000; Hunter and Ruffner, 2001; Walker *et al*., 2021).

The vines did not show the expected response, i.e., higher water use efficiency, during the active canopy period (approximately until véraison), which is normally driven by a better canopy microclimate (Hunter and Visser, 1988). A more open canopy would naturally lead to higher leaf activity unless it is too open, leaf senescence and photo-inhibition occur, and water stress prevails (Hunter and Visser, 1989, and references therein; Hunter, 1998b). The abiotic environment (such as high temperatures and drier conditions) and the gradual ageing of the leaves as grape ripening proceeded ostensibly reduced its water use efficiency, despite the positive radiation and increasing sink demand for sucrose production during this period, e.g., for transport to and accumulation in berries (Coombe, 1992; Hunter *et al*., 1994; Hunter *et al*., 2014; Carlomagno *et al*., 2018). Synergistic effects of water stress, intense radiation and high temperature on photo-inhibition have previously been shown for the grapevine (Schultz, 1996; Medrano *et al*., 2015). Protective mechanisms against photo-oxidation that may be triggered in leaves to avoid down-regulation of photosynthesis under stressful conditions include increases in superoxide radical production, H_2O_2 concentration, anti-oxidant enzyme activity (e.g., superoxide dismutase, ascorbate peroxidase, dihydro-ascorbate reductase, catalase), the osmotic stress-related amino acid proline, the hormone abscisic acid, and carotenoids (involvement of carotenes, like ß-carotene, and xanthophylls, like the epoxide-free lutein and zeaxanthin and the di-epoxide violaxanthin, in the dissipation of harmful excess excitation energy), all accompanied by a decrease in the chlorophyll:carotenoid ratio (Demmig-Adams *et al*., 1996; Wehner *et al*., 2004; Nenko *et al*., 2021). The ability to produce the carotenoid zeaxanthin in response to light-mediated environmental stress may serve as an indicator of a plant's ability to endure terroir-related conditions (Young, 1991).

Photo-inhibition may not always be dynamic (reversible/recoverable), but chronic photo-inhibition may occur, the latter being the permanent loss of functionality of the photosystem II reaction centre, leading to chlorosis and necrosis (Long *et al*., 1994). Many factors and processes that are important for optimal physiological performance, such as water status, light microclimate, ambient temperature, canopy activity, carbon distribution, sink demand, source supply, reserve starch levels, etc., may affect photo-inhibition vulnerability and may counterbalance each other and change net physiological activity and supply to reproductive and reserve organs of vines. Thus, although the general effect of photo-inhibition may convert into decreased efficiency and crop dry matter loss (Long *et al*., 1994), the reversal may not necessarily lead to carbon gain at the field level, as it would require all cultivation practices to allow optimal vine metabolic processes under the demands of abiotic and biotic factors. Moreover, whilst the lower water potential of wider-spaced vines may have had an effect on stomatal behaviour and photosynthetic activity, a myriad of other physiological responses to vine structure expansion with wider spacing might have played a role, such as unequal development of aboveground and subterranean growth and thus disturbance in the capacity of supply and demand bodies; distances of translocation may have deleteriously affected flow and volume via support channels; crop load per vine; higher water (irrigation) and nutrition (fertilisation) requirements/demands, etc. Furthermore, although a plausible deduction from the diurnal accumulation of carbohydrates (Chaumont *et al*., 1994; Hunter *et al*., 1994), it remains difficult to validate source:sink feedback mechanisms under field conditions in a complex multi-organ disposition such as the grapevine where carbon is partitioned between photosynthesising, reproductive and perennial storage tissue under the influence of manipulation (Chaumont *et al*., 1994; Hunter *et al*., 1995b) and even in other plants under greenhouse conditions where end-product inhibition of photosynthesis is expected (Goldschmidt and Huber, 1992).

Berry temperature generally increased with wider vine spacing (Figure 9). Berry temperature differences between the different bunch positions (canopy exterior and interior) were more pronounced during post- than during pre-véraison, most likely because the canopy was fully developed and settled and provided more protection to interior-exposed berries at the time of measurement during post-véraison. The berry temperature changed according to the radiation of the canopy wall, whereas the bunches carried in the interior of the canopy were largely protected from the

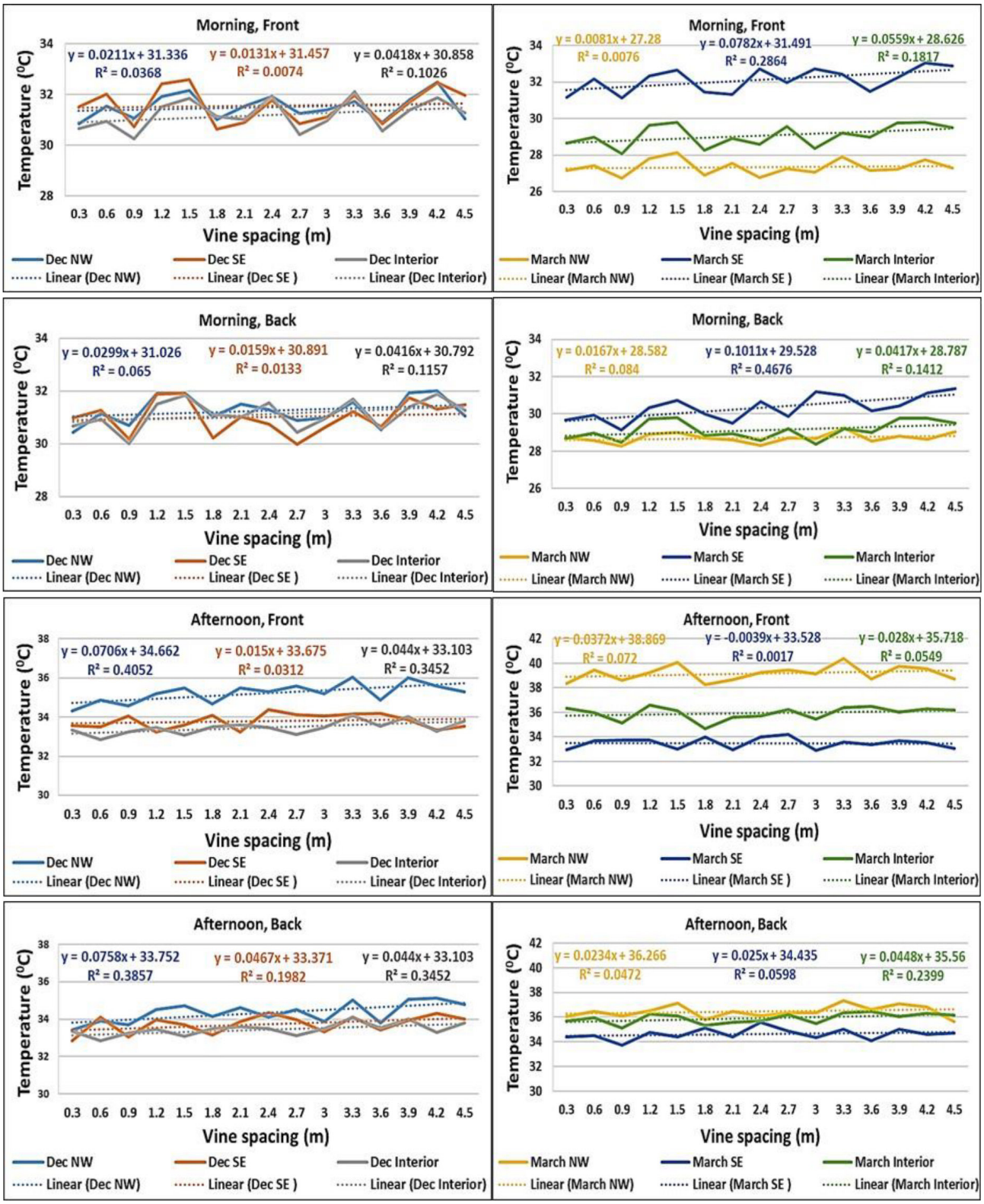


FIGURE 9. Grape berry temperature measured in the morning and in the afternoon on bunches on the exterior/front of different canopy sides and in the interior/middle of the canopy [(pre-véraison (left) and post-véraison (right)] of NNE-SSW oriented Shiraz/101-14 Mgt (means of 2017–2020 seasons).

radiation impact. This was also found in a study that detailed berry temperature differences as affected by vineyard row orientation (Hunter *et al.*, 2021). Collectively, the berry pulp temperatures were much higher in the afternoon than in the morning, irrespective of position. At the respective times of measurement pre- and post-véraison, berry temperatures fell within and even far beyond those that are considered suppressive for anthocyanin accumulation (Hunter and Bonnardot, 2011; Matsuda *et al.*, 2021), more so the wider the vines were spaced. An increase in pulp

temperature, along with an increase in light exposure of the berries and a decrease in ripeness level with wider spacing, presented an interesting scenario, but clearly resulted from a whole-plant stress condition. As found by Hunter *et al*. (2021), successively experienced excessive light exposure, and heating of the grapes seemed not conducive to high-quality grape composition. In the latter study, clear differences in basic berry composition and individual anthocyanins according to exposure and pulp temperature were shown.

4. General

For closer-spaced vines, lower leaf water potential values have been expected with the limited soil volume availability and higher root density that may have increased localised water depletion, especially under high ambient temperature conditions. Instead, vines apparently showed tighter stomatal behaviour to reduce transpirational water loss or as influenced by a denser canopy. Not excluded is a reduction in vessel diameter of shoot basal internodia and the vulnerability to embolism formation in shoots and especially roots, which would have reduced hydraulic conductance and leaf water availability/transpiration and may have affected vegetative growth in general (Lovisolo and Schubert, 1998; Lovisolo *et al*., 2008). A denser canopy, as indicated by the canopy light conditions, may have had a restrictive effect on leaf activity (Smart *et al*., 1990; Hunter, 2000). The grapevine canopy mostly comprises a combination of 1) leaves in shady positions that would have a high need for efficient light collection and 2) fully exposed leaves that would have a high need for photoprotection. In the context of this study, canopies of the narrow spacing treatments had a higher ratio of shaded:exposed leaves and canopies of the wide spacing treatments had the opposite, thereby, respectively, having lower and higher effective leaf area per surface (data not shown in this paper). However, it seems that at least for the widest spacing treatments, partial or total canopy photoprotection was required, and, thus, leaf efficiency may have been affected. Nonetheless, the grapevine canopy would have to have sufficient flexibility under any circumstances, including cloudy versus high-intensity sunlight conditions, to change the carotenoid composition according to radiation intensity. A determining factor for the speed of recovery from either period for the different spacing treatments would be the physiological status, as affected by levels of other biotic and abiotic stress-inducing parameters, i.e., environmental (e.g., water stress) and/or cultivation practices (e.g., the sensitivity of the variety-rootstock combination; depth and composition of the root system; canopy management) (Hunter *et al*., 2016a). The same parameters would be applicable to avoid imbalances altogether and allow optimal physiological functioning as well as physical protection of leaves (and grapes) in the canopy.

Both root system and canopy are involved in water potential and photosynthetic activity regulation, and the interaction between these two plant departments may lead to a lack of (expected) differences. Vine spacing affects both aboveground and belowground departments of the vine and, therefore, induces an array of complex interactive physiological trigger and homeostatic mechanisms. If these departments are not developing in a balanced way, the reaction of the grapevine to environmental and cultivation practices would be disturbed, often leading to erratic, unpredictable, underperforming, and unsustainable behaviour. Such a condition would thus not be conducive to a long lifespan and tolerance of the grapevine to environmental challenges, such as those associated with a change in climate.

5. Guidelines

Under the conditions of this study and according to all data at hand, the following guidelines surfaced:

• Vine spacing had a steering effect on the development of the root system, affecting root generation and distribution and the potential for soil exploration and utilisation.

• The positive relationship between aboveground and subterranean growth was confirmed.

• Root density in the fine, extension and permanent categories increased with narrower spacing, while deeper soil layers were explored by the roots. This increased the vertical root preferential zone and total number of roots in the soil profile; lateral distribution appeared uniform over the allocated distances.

• Fine roots dominated the soil profiles across spacing treatments, demonstrating their importance for soil utilisation.

• Colonisation of deeper soil layers might have allowed the deep penetrating roots to utilise water and minerals in soil layers that would not have been available if roots stayed in shallower soil layers with wide vine spacing.

• Results validated the hypothesis that more fertile soil conditions with minimum depth obstructive barriers and other soil physical and chemical restrictions, adequate oxygen supply, sufficient water retainment, and especially good drainage would stimulate roots to explore deeper soil layers to compensate for lateral space restriction and aerial growth demand; this would be well aided by pre-establishment deep soil preparation (especially in soils that have root prohibitive layers).

• A deep penetrating and genetically more drought-resistant root system would increase the buffer potential and resilience of the vine against adverse climatic conditions, such as high temperature, low humidity, low rainfall, strong winds, heat waves, etc., while providing a higher natural capacity for consistency in growth, production, and grape quality (vineyard sustainability).

• A global rise in temperature (and most likely regional drought) puts increasing emphasis on creating a healthy root system that colonises deeper soil layers and, thus, a deeper preferential root zone. This would most presumably also develop naturally with time as the ambient temperature increases, and the longer period of exposure to heat from season to season results in warmer and drier topsoil below the vineyard rows, thereby pushing the roots deeper into cooler and moister zones.

• Phenological stage management actions, irrigation, and fertilisation should be synchronised with root activity peaks to support perseverance in growth and grape ripening.

• Optimum vine spacing appeared to be 1.8 m, i.e., approx. 4 m^2 surface area/vine or 2525 vines/ha.

• Both closer and wider spacing than 1.8 m induced limitations in physiological activity, i.e., stressful conditions affecting growth, microclimate, leaf function and berry temperature, while decreasing yield capacity.

• The physiological status and berry exposure and pulp temperature differences that occurred would have affected primary and secondary berry composition amongst vine spacing treatments, with implications for wine quality.

The ultimate objective to fully utilise an allocated land surface/soil volume for grape production is complicated by a magnitude of abiotic and biotic factors as well as vineyard practices that may introduce antagonistic effects on aboveground and belowground growth environments. All elements need to be considered for each specific establishment.

CONCLUSION

In this study, vine reaction to aboveground and belowground spaces was monitored. Carbon allocation differences among vine spacing treatments were evident in root system density and depth. Trends found for root density, canopy microclimate, water potential, photosynthesis, and efficiency of water use seemed to indicate that widely spaced vines experienced development-originating physiological restrictions and more stress. Confinement and progressive extension of the grapevine structure led to a gradual transition from a higher vigour (perhaps mostly root growth/physiology induced) to a lower vigour (perhaps mostly structure and canopy/physiology induced) condition. A complex interplay between vine structure enlargement, root formation and distribution, canopy physiological parameters, and carbon distribution occurred. Clearly, the drought tolerance, size and activity of the root system, together with maintenance of turgor, to guarantee uninterrupted mineral and assimilate supply to the various demand points in the grapevine, would be critical for productive survival. This study provided information at practical and scientific levels to aid strategic approaches towards a solid foundation for full utilisation and expression of the specific terroir conditions and sustainable growth and production.

ACKNOWLEDGEMENTS

Agricultural Research Council and SA Wine Industry (through Winetech) for funding. Our gratitude goes to the Viticulture Department (L.F. Adams, A. Marais) and ARC Robertson Farm personnel for technical assistance.

REFERENCES

Archer, E, & Hunter, J.J. (2004). Vine balance: Its importance to successful cultivation. *Wineland*, 2, 60-66.

Archer, E, & Saayman, D. (2018). Wingerdwortels. African Sun Media, Stellenbosch, South Africa. 229 pp. https://doi.org/10.18820/9781928314370

Archer, E, & Strauss, H.C. (1985). The effect of plant density on root distribution of three-year-old grafted 99 Richter grapevines. *South African Journal of Enology and Viticulture*, 6, 25-30. https://doi.org/10.21548/6-2-2347

Archer, E, & Strauss, H.C. (1989). The effect of plant spacing on the water status of soil and grapevines. *South African Journal of Enology and Viticulture*, 10, 49-58. https://doi.org/10.21548/10-2-2286

Archer, E, Swanepoel, J.J, & Strauss, H.C. (1988). Effect of plant spacing and trellising systems on grapevine root distribution. In: The grapevine root and its environment (Compiler: J. Van Zyl). Dept Agriculture and Water Supply, Pretoria, South Africa.

Böhm, W. (1979). Methods of studying root systems. Ecological studies. Vol. 33, Springer Verlag, Berlin. https://doi.org/10.1007/978-3-642-67282-8_2

Candolfi-Vasconcelos, M.C, Candolfi, M.P, & Koblet, W. (1994). Retranslocation of carbon reserves from the woody storage tissues into the fruit as a response to defoliation stress during the ripening period in *Vitis vinifera* L. *Planta*, 192, 567-573. https://doi.org/10.1007/BF00203595

Carlomagno, A, Novello, V, Ferrandino, A, Genre, A, Lovisolo, C. & Hunter, J.J. (2018). Pre-harvest berry shrinkage in cv 'Shiraz' (*Vitis vinifera* L.): understanding sap flow by means of tracing. *Scientia Horticulturae*, 233, 394-406. https://doi.org/10.1016/j.scienta.2018.02.014

Champagnol, F. (1979). La densité de plantation en viticulture. *Progrés Agric. Vitic*., 96, 185-195.

Chaumont, M, Morot-Gaudry, J-F, & Foyer, C.H. (1994). Seasonal and diurnal changes in photosynthesis and carbon partitioning in *Vitis vinifera* leaves in vines with and without fruit. *Journal of Experimental Botany*, 45, 1235-1243. https://doi.org/10.1093/jxb/45.9.1235

Conradie, W.J. (1983). Liming and choice of rootstocks as cultural techniques for vines in acid soils. *South African Journal of Enology and Viticulture*, 4, 39-44. https://doi.org/10.21548/4-2-2368

Coombe, B.G. (1992). Research on development and ripening of the grape berry. *American Journal of Enology and Viticulture*, 43, 101-110. https://doi.org/10.5344/ajev.1992.43.1.101

Davies, W.J, Metcalfe, J, Lodge, T.A, & Da Costa, A.R. (1986). Plant growth substances and the regulation of growth under drought. *Australian journal of plant physiology*, 13, 105-125. https://doi.org/10.1071/PP9860105

Demmig-Adams, B, Gilmore, A.M, & Adams, W.W. (1996). *In vivo* functions of carotenoids in higher plants. *FASEB J*., 10, 403-412. https://doi.org/10.1096/fasebj.10.4.8647339

Düring, H. (1991). Determination of the photosynthetic capacity of grapevine leaves. *Vitis*, 30, 49-56.

Edson, C.E, Howell, G.S, & Flore, J.A. (1995). Influence of crop load on photosynthesis and dry matter partitioning of Seyval grapevines. III. Seasonal changes in partitioning, vine morphology, yield, and fruit composition. *American Journal of Enology and Viticulture*, 46, 478-485. https://doi.org/10.5344/ajev.1995.46.4.478

Epstein, E, & Bloom, A.J. (2005) (2nd Ed). Mineral Nutrition of Plants: Principles and Perspectives. Sinauer, Underland, MA.

Escalona, J.M, Tomàs, M, Martorell, S, Medrano, H, Ribas-Carbo, M, & Flexas, J. (2012). Carbon balance in grapevines under different soil water supply: importance of whole plant respiration. *Australian Journal of Grape and Wine Research*, 18, 308-318. https://doi.org/10.1111/j.1755-0238.2012.00193.x

Feng, Y.L, Cao, K.F, & Feng, Z.L. (2002). Thermal dissipation, leaf rolling and inactivation of PS II reaction centres in *Amomum villosum*. *Journal of Tropical Ecology*, 18, 865-876. https://doi.org/10.1017/S0266467402002560

Field, S.K, Smith, J.P, Holzapfel, B.P, Hardie, W.J, & Emery, R.J.N. (2009). Grapevine response to soil temperature: Xylem cytokinins and carbohydrate reserve mobilization from budbreak to anthesis. *American Journal of Enology and Viticulture*, 60, 164-172. https://doi.org/10.5344/ajev.2009.60.2.164

Fischer, R.A, & Turner, N.C. (1978). Plant productivity in the arid and semiarid zones. *Annual Review of Plant Physiology*, 29, 277-317. https://doi.org/10.1146/annurev.pp.29.060178.001425

Flexas, J, Bota, J, Escalona, J.M, Sampol, B, & Medrano, H. (2002). Effects of drought on photosynthesis in grapevines under field conditions: an evaluation of stomatal and mesophyll limitations. *Funct. Plant Biology*, 29, 461-471. https://doi.org/10.1071/PP01119

Freeman, B.M. (1983). At the root of the vine. *Austr. Grapegrower Winemaker*, 4, 58-64.

Gambetta, G.A, Fei, J, Rost, T.L, Knipfer, T, Matthews, M.A, Shackel, K.A, Walker, M.A, & McElrone, A.J. (2013). Water uptake along the length of grapevine fine roots: developmental anatomy, tissue-specific aquaporin expression, and pathways of water transport. *Plant Physiol*., 163, 1254-1265. https://doi.org/10.1104/pp.113.221283

Gambetta, G.A, Herrera, J.C, Dayer, S, Feng, Q, Hochberg, U, & Castellarin, S.D. (2020). The physiology of drought stress in grapevine: towards an integrative definition of drought tolerance. *Journal of Experimental Botany*, 71, 4658-4676. https://doi.org/10.1093/jxb/eraa245

Gamon, J.A, & Pearcy, R.W. (1989). Leaf movement, stress avoidance and photosynthesis in *Vitis californica*. *Oecologia*, 79, 475-481. https://doi.org/10.1007/BF00378664

Giaquinta, R.T. (1983). Phloem loading of sucrose. *Annual Review of Plant Physiology*, 34, 347-387. https://doi.org/10.1146/annurev.pp.34.060183.002023

Goldschmidt, E.E, & Huber, S.C. (1992). Regulation of Photosynthesis by end-product accumulation in leaves of plants storing starch, sucrose, and hexose sugars. *Plant Physiol*., 99, 1443-1448. https://doi.org/10.1104/pp.99.4.1443

Greer, D, & Rogiers, S.Y. (2009). Water flux of *Vitis vinifera* L. cv. Shiraz bunches throughout development and in relation to late-season weight loss. *American Journal of Enology and Viticulture*, 60, 155-163. https://doi.org/10.5344/ajev.2009.60.2.155

Guan, X.Q, Zhao, S.J, Li, D.Q, & Shu, H.R. (2004). Photoprotective function of photorespiration in several grapevine cultivars under drought stress. *Photosynthetica*, 42, 31-36. https://doi.org/10.1023/B:PHOT.0000040566.55149.52

Hochberg, U, Degu, A, Toubiana, D, Gendler, T, Nikoloski, Z, Rachmilevitch, S, & Fait, A. (2013). Metabolite profiling and network analysis reveal coordinated changes in grapevine water stress response. *BMC Plant Biol*., 13, 184. https://doi.org/10.1186/1471-2229-13-184

Hochberg, U, Rockwell, F.E, Holbrook, N.M, & Cochard, H. (2018). Iso/anisohydry: a plant-environment interaction rather than a simple hydraulic trait. *Trends in Plant Science*, 23, 112-120. https://doi.org/10.1016/j.tplants.2017.11.002

Hochberg, U., Windt, C.W, Ponomarenko, A, Zhang, Y-J, Gersony, J, Rockwell, F.E, & Holbrook, N.M. (2017). Stomatal closure, basal leaf embolism, and shedding protect the hydraulic integrity of grape stems. *Plant Physiology*, 174, 764-775. https://doi.org/10.1104/pp.16.01816

Hudson, D. (1966). Fitting segmented curves whose join points have to be estimated. *Journal of the American Statistical Association*, 61, 1097-1129. https://doi.org/10.1080/01621459.1966.10482198

Hunter, J.J. (1998a). Plant spacing implications for grafted grapevine I. Soil characteristics, root growth, dry matter partitioning, dry matter composition and soil utilisation. *South African Journal of Enology and Viticulture*, 19, 25-34. https://doi.org/10.21548/19-2-2242

Hunter, J.J. (1998b). Plant spacing implications for grafted grapevine II. Soil water, plant water relations, canopy physiology, vegetative and reproductive characteristics, grape composition, wine quality and labour requirements. *South African Journal of Enology and Viticulture*, 19, 35-51. https://doi.org/10.21548/19-2-2243

Hunter, J.J. (2000). Implications of seasonal canopy management and growth compensation in grapevine. S. Afr. J. Enol. Vitic., 21, 81-91. https://doi.org/10.21548/21-2-2215

Hunter, J.J, Archer, E, Van Schalkwyk, D, Strever, A.E, & Volschenk, C.G. (2016a). Grapevine roots: interaction with natural factors and agronomic practices. *Acta Hortic*., 1136, 63-80. https:/doi.org/10.17660/ActaHortic.2016.1136.10

Hunter, J.J, Archer, E, & Volschenk, C.G. (2010). Vineyard management for environment valorisation. Proc. VIIIth Int. Terroir Zoning Congress, 14-18 June, Soave, Italy, 7-3-7-15.

Hunter, J.J, & Bonnardot, V. (2011). Methodology to assess vine cultivation suitability of South African regions using optimum climatic ranges for key physiological processes. *South African Journal of Enology and Viticulture*, 32, 137-154.

Hunter, J.J, & Le Roux, D.J. (1992). The effect of partial defoliation on development and distribution of roots of *Vitis vinifera* L. cv. Cabernet Sauvignon grafted onto rootstock 99 Richter. *American Journal of Enology and Viticulture*, 43, 71-78. https://doi.org/10.5344/ajev.1992.43.1.71

Hunter, J.J, & Ruffner, H.P. (2001). Assimilate transport in grapevines - effect of phloem disruption. *Australian Journal of Grape and Wine Research*, 7, 118-126. https://doi.org/10.1111/j.1755-0238.2001.tb00198.x

Hunter, J.J, Ruffner, H.P, & Volschenk, C.G. (1995a). Starch concentrations in grapevine leaves, berries and roots and the effect of canopy management. *South African Journal of Enology and Viticulture*, 16, 35-40. https://doi.org/10.21548/16-2-2270

Hunter, J.J, Ruffner, H.P, Volschenk, C.G, & Le Roux, D.J. (1995b). Partial defoliation of *Vitis vinifera* L. cv. Cabernet Sauvignon/99 Richter: Effect on root growth, canopy efficiency, grape composition and wine quality. *American Journal of Enology and Viticulture*, 46, 306-314. https://doi.org/10.5344/ajev.1995.46.3.306

Hunter, J.J, Skrivan, R, & Ruffner, H.P. (1994). Diurnal and seasonal physiological changes in leaves of *Vitis vinifera* L.: CO_2 assimilation rates, sugar levels and sucrolytic enzyme activity. *Vitis*, 33, 189-195.

Hunter, J.J, Tarricone, L, Volschenk, C.G, Giacalone, C, Melo, M.S, & Zorer, R. (2020). Grapevine physiological response to row orientation-induced spatial radiation and microclimate changes. *OENO One*, 54, 411-433. https://doi.org/10.20870/oeno-one.2020.54.2.3100

Hunter, J.J, & Visser, J.H. (1988). Distribution of 14C-photosynthetate in the shoot of Vitis vinifera L. cv. Cabernet Sauvignon. II. The

effect of partial defoliation. *South African Journal of Enology and Viticulture*, 9, 10-5. https://doi.org/10.21548/9-1-2303

Hunter, J.J, & Visser, J.H. (1989). The effect of partial defoliation, leaf position and developmental stage of the vine on leaf chlorophyll concentration in relation to the photosynthetic activity and light intensity in the canopy of *Vitis vinifera* L. cv. Cabernet Sauvignon. *South African Journal of Enology and Viticulture*, 10, 67-73. https://doi.org/10.21548/10-2-2289

Hunter, J.J, Volschenk, C.G, Mania, E, Vicente Castro, A, Booyse, M, Guidoni, S, Pisciotta, A, Di Lorenzo, R, Novello, V, & Zorer, R. (2021). Grapevine row orientation mediated temporal and cumulative microclimatic effects on grape berry temperature and composition. *Agricultural and Forest Meteorology*, 310. https://doi.org/10.1016/j.agrformet.2021.108660

Hunter, J.J, Volschenk, C.G, Novello, V, Strever, A.E, & Fouché, G.W. (2014). Integrative effects of vine water relations and grape ripeness level of *Vitis vinifera* L. cv. Shiraz/Richter 99. I. Physiological changes and vegetative-reproductive growth balances. *South African Journal of Enology and Viticulture*, 35, 332-358. https://doi.org/10.21548/35-2-1021

Hunter, J.J, Volschenk, C.G, & Zorer, R. (2016b). Vineyard row orientation of *Vitis vinifera* L. cv. Shiraz/101-14 Mgt: Climatic profiles and vine physiological status. *Agricultural and Forest Meteorology*, 228, 104-119. https://doi.org/10.1016/j.agrformet.2016.06.013

Kriedemann, P.E. (1968). Photosynthesis in vine leaves as a function of light intensity, temperature, and leaf age. *Vitis*, 7, 213-220.

Kudoyarova, G, Arkhipova, T, Korshunova, T, Bakaeva, M, Loginov, O, & Dodd, I.C. (2019). Phytohormone mediation of interactions between plants and non-symbiotic growth promoting bacteria under edaphic stresses. *Frontiers in Plant Science*, 10. https://doi.org/10.3389/fpls.2019.01368

Levene, H. (1960). "Robust test in the equality of variance" in Contributions to Probability and Statistics. Iolkin Palo Alto (Eds), CA, Stanford University Press, 278-292.

Levitt, J. (1980). Responses of plant to environmental stresses, vol II. Academic Press, New York.

Linares Torres, R, De la Fuente Lloreda, M, Junquera Gonzalez, P, Lissarrague García-Gutierrez, J.R, & Baeza Trujillo, P. (2018). Effect of soil management strategies on the characteristics of the grapevine root system in irrigated vineyards under semi-arid conditions. *Australian Journal of Grape and Wine Research*, 24, 439-449. https://doi.org/10.1111/ajgw.12359

Long, S.P, Humphries, S, & Falkowski, P.G. (1994). Photoinhibition of photosynthesis in nature. *Annual Review of Plant Physiology and Plant Molecular Biology*, 45, 633-662. https://doi.org/10.1146/annurev.pp.45.060194.003221

Lovisolo, C, Perrone, I, Carra, A, Ferrandino, A, Flexas, J, Medrano, H. & Schubert, A. (2010). Drought-induced changes in development and function of grapevine (*Vitis spp*.) organs and in their hydraulic and non-hydraulic interactions at the whole-plant level: a physiological and molecular update. *Functional Plant Biology*, 37, 98-116. https://doi.org/10.1071/FP09191

Lovisolo, C, Perrone, I, Hartung, W, & Schubert, A. (2008). An abscisic acid related reduced transpiration promotes gradual embolism repair when grapevines are rehydrated after drought. *New Phytol*ogist, 180, 642-651. https://doi.org/10.1111/j.1469-8137.2008.02592.x

Lovisolo, C, & Schubert, A. (1998). Effects of water stress on vessel size and xylem hydraulic conductivity in *Vitis vinifera* L. *Journal of Experimental Botany*, 49, 693-700. https://doi.org/10.1093/jexbot/49.321.693

Matsuda, K, Gao-Takai, M, Date, A, Suzuki, T. (2021). Suppression of red color development associated with anthocyanin accumulation in the epicarp of grape (*Vitis labrusca* x *vinifera* cv. Ruby Roman) berries caused by air temperature in daylight periods higher than 33^0C during maturation. *Scientia Horticulturae*, 288, 1-7. https://doi.org/10.1016/j.scienta.2021.110381

Medrano, H, Tomás, M, Martorell, S, Escalona, J.M, Pou, A, Fuentes, S, Flexas, J, & Bota, J. (2015). Improving water use efficiency of vineyards in semi-arid regions. A review. *Agronomy for Sustainable Development*, 35, 499-517. https://doi.org/10.1007/s13593-014-0280-z

Mohr, H.D. (1996). Periodicity of root tip growth of vines in the Moselle valley. *Vitic. Enol. Sci*., 51, 83-90.

Nagarajah, S. (1987). Effects of soil texture on the rooting patterns of Thompson Seedless vines on own roots and on Ramsey rootstock in irrigated vineyards. *American Journal of Enology and Viticulture*, 38, 54-59. https://doi.org/10.5344/ajev.1987.38.1.54

Naor, A, & Wample, R.L. (1996). Diurnal internode growth rate in field-grown 'Concord' (*Vitis labrusca*) grapevines. *Journal of Horticultural Sciences*, 71, 167-171. https://doi.org/10.1080/14620316.1996.11515393

Nenko, N, Kiseleva, G, Ilina, I, Sokolova, V, & Zaporozhets, N. (2021). Grapes adaptive resistance to summer stresses in the conditions of climate change. *E3S Web of Conferences*, 254, 02019. https://doi.org/10.1051/e3sconf/202125402019

Ott, R.L, & Longnecker, M. (2010) (6th Ed). An Introduction to Statistical methods and data analysis. Belmont, CA, Duxbury Press, 1152 pp.

Overvoorde, P, Fukaki, H, & Beeckman, T. (2010). Auxin control of root development. Cold Spring Harb. *Perspectives in Biolog*, 2, a001537. https://doi.org/10.1101/cshperspect.a001537

Patakas, A, & Noitsakis, B. (1999). Mechanisms involved in diurnal changes of osmotic potential in grapevines under drought conditions. *Journal of Plant Physiology*, 154, 767-774. https://doi.org/10.1016/S0176-1617(99)80256-9

Patrick, J.W. (1997). Phloem unloading: Sieve element unloading and post-sieve element transport. *Annual Review of Plant Physiology and Plant Molecular Biology*, 48, 191-222. https://doi.org/10.1146/annurev.arplant.48.1.191

Pisciotta, A, Di Lorenzo, R, Volschenk, C.G, & Hunter, J.J. (2011). Relation between row and leaf orientation of Syrah/101-14 Mgt. In: Proc. 17th Int. Symp. GiESCO, Asti and Alba (CN), Italy, 29 Aug-2 Sept, 555-558.

Poni, S, Intrieri, C, & Silvestroni, O. (1994). Interactions of leaf age, fruiting, and exogenous cytokinins in Sangiovese grapevines under non-irrigated conditions. I. Gas exchange. *American Journal of Enology and Viticulture*, 45, 71-78. https://doi.org/10.5344/ajev.1994.45.1.71

Pou, A, Medrano, I, Tomàs, M, Martorell, S, Ribas-Carbó, M, & Flexas, J. (2012). Anisohydric behaviour in grapevines results in better performance under moderate water stress and recovery than isohydric behaviour. *Plant Soil*, 359, 335-349. https://doi.org/10.1007/s11104-012-1206-7

Richards, D. (1983). The grape root system. *Horticultural Reviews*, 5, 127-168. https://doi.org/10.1002/9781118060728.ch3

Rivero, R.M, Kojima, M, Gepstein, A, Sakakibara, H, Mittler, R, Gepstein, S, & Blumwald, E. (2007). Delayed leaf senescence induces extreme drought tolerance in a flowering plant. *PNAS*, 104, 19631-19636. https://doi.org/10.1073/pnas.0709453104

Rodrigues, M.L, Chaves, M.M, Wendler, R, David, M.M, Quick, W.P, Leegood, R.C, Stitt, M, & Pereira, J.S. (1993). Osmotic

adjustment in water stressed grapevine leaves in relation to carbon assimilation. *Australian journal of plant physiology*, 20, 309-321. https://doi.org/10.1071/PP9930309

Saayman, D. (1982). Soil preparation studies: II. The effect of depth and method of soil preparation and of organic material on the performance of *Vitis vinifera* (var. Colombar) on Clovelly/Hutton soil. *South African Journal of Enology and Viticulture*, 3, 61-74. https://doi.org/10.21548/3-2-2383

Santos, J.A, Fraga, H, Malheiro, A.C, Moutinho-Pereira, J, Dinis, L.T., Correia C., Moriondo M., Leolini, L, Dibari, C, Costafreda-Aumedes, S, Kartschall, T, Menz, C, Molitor, D, Junk, J, Beyer, M, & Schultz, H.R. (2020). A review of the potential climate change impacts and adaptation options for European viticulture. *Applied Sciences*, 10, 3092. https://doi.org/10.3390/app10093092

Scholander, P.F, Hammel, H.T, Bradstreet, E.D, & Hemmingsen, E.A. (1965). Sap pressure in vascular plants. *Science*, 148, 339-346. https://doi.org/10.1126/science.148.3668.339

Schultz, H.R. (1996). Water relations and photosynthetic responses of two grapevine cultivars of different geographical origin during water stress. *Acta Horticulturae*, 427, 251-266. https://doi.org/10.17660/ActaHortic.1996.427.30

Schultz, H.R, & Matthews, M.A. (1993). Xylem development and hydraulic conductance in sun and shade shoots of grapevine (*Vitis vinifera* L.): evidence that low light uncouples water transport capacity from leaf area. *Planta*, 190, 393-406. https://doi.org/10.1007/BF00196969

Shapiro, S.S, & Wilk, M.B. (1965). An analysis of variance test for normality (complete samples). *Biometrika*, 52, 591-611. https://doi.org/10.1093/biomet/52.3-4.591

Slavtcheva, T, & Pourtchev, P. (2003). Root system development with cv. Dimiat grapevines planted on leached chernozem in relation to the pruning. *Pochvoznanie, Agrokhimiya i-Ekologiya*, 38, 49-53.

Smart, R.E, Dick, J.K, Gravett, I.M, & Fisher, B.M. (1990). Canopy management to improve grape yield and wine quality - principles and practices. *South African Journal of Enology and Viticulture*, 11, 3-17. https://doi.org/10.21548/11-1-2232

Southey, J.M. (1992). Grapevine rootstock performance under diverse conditions in South Africa. In: Wolpert J.A., Walker M.A. & Weber E. (eds.). Proc. Rootstock Seminar: A worldwide Perspective, 24 June 1992, Reno, Nevada, USA, 27-51.

Stoll, M, Loveys, B, & Dry, P. (2000). Hormonal changes induced by partial root zone drying of irrigated grapevine. *Journal of Experimental Botany*, 51, 1627-1634. https://doi.org/10.1093/jexbot/51.350.1627

Swanepoel, J.J, & Southey, J.M. (1989). The influence of rootstock on the rooting pattern of the grapevine. *South African Journal of Enology and Viticulture*, 10, 23-28. https://doi.org/10.21548/10-1-2295

Vandeleur, R.K, Mayo, G, Shelden, M.C, Gilliham, M, Kaiser, B.N, & Tyerman, S.D. (2009). The role of plasma membrane intrinsic protein aquaporins in water transport through roots: diurnal and drought stress responses reveal different strategies between isohydric and anisohydric cultivars of grapevine. *Plant Physiology*, 2009, 445-460. https://doi.org/10.1104/pp.108.128645

Van Huyssteen, L, & Weber, H.W. (1980). The effect of selected minimum and conventional tillage practices in vineyard cultivation on vine performance. *South African Journal of Enology and Viticulture*, 1, 77-83. https://doi.org/10.21548/1-2-2416

van Leeuwen, C., & Destrac-Irvine, A. (2017). Modified grape composition under climate change conditions requires adaptations in the vineyard. *OENO One*, 51. https://doi.org/10.20870/oeno-one.2017.51.2.1647

van Leeuwen, C, Pieri, P, Gowdy, M., Ollat, N., & Roby, J-P. (2019). Reduced density is an environmental friendly and cost effective solution to increase resilience to drought in vineyards in a context of climate change. *OENO One*, 53. https://doi.org/10.20870/oeno-one.2019.53.2.2420

Van Zyl, J.L. (1984). Response of Colombar grapevines as regards quality aspects and growth. *South African Journal of Enology and Viticulture*, 5, 19-28. https://doi.org/10.21548/5-1-2365

Van Zyl, J.L, & Weber, H.W. (1981). The effect of various supplementary irrigation treatments on plant and soil moisture relationships in a vineyard (*Vitis vinifera* var. Chenin blanc). *South African Journal of Enology and Viticulture*, 2, 83-99. https://doi.org/10.21548/2-2-2400

Walker, R.P, Chen, Z-H, & Famiani, F. (2021). Gluconeogenesis in Plants: A key interface between organic acid/amino acid/lipid and sugar metabolism. *Molecules*, 26, 1-18. https://doi.org/10.3390/molecules26175129

Wehner, A, Storf, S, Jahns, P, & Schmid, V.H.R. (2004). De-epoxidation of violaxanthin in light-harvesting complex I proteins. *Journal of Biological Chemistry*., 279, 26823-26829. https://doi.org/10.1074/jbc.M402399200

Werner, C, Correia, O, & Beyschlag, W. (1999). Two different strategies of Mediterranean macchia plants to avoid photoinhibitory damage by excessive radiation levels during summer drought. *Acta Oecologica*, 20, 15-23. https://doi.org/10.1016/S1146-609X(99)80011-3

Yang, J, Zhang, J, Wang, Z, Zhu, Q, & Liu, L. (2002). Abscisic acid and cytokinins in the root exudates and leaves and their relationship to senescence and remobilization of carbon reserves in rice subjected to water stress during grain filling. *Planta*, 2002, 645-652. https://doi.org/10.1007/s00425-002-0789-2

Young, A.J. (1991). The photoprotective role of carotenoids in higher plants. *Physiologia Plantarum*, 83, 702-708. https://doi.org/10.1111/j.1399-3054.1991.tb02490.x

Zimmermann, M.H. (1983). Xylem structure and the ascent of sap. Berlin, *Springer*. https://doi.org/10.1007/978-3-662-22627-8

Zufferey, V. (2016). Leaf respiration in grapevine (*Vitis vinifera* 'Chasselas') in relation to environmental and plant factors. *Vitis*, 55, 65-72.