Scion–rootstock interactions influence the growth and behaviour of the grapevine root system in a heavy clay soil

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Abstract

Background and Aims: Generally, grapevine roots have been less studied than the above-ground parts of the plant. Here we analyse scion–rootstock interactions in mature vines growing in a heavy clay soil in a climate characterised by severe summer drought to investigate the effect of the scion–rootstock interaction in a suboptimal soil.

Methods and Results: The rootstocks, 34 Ecole de Montpellier, 140 Ruggeri and 1103 Paulsen, were grafted onto Nerello Mascalese and Nero d'Avola scions and assessed along with self-rooted vines. Root distribution and root architecture were analysed using the profile wall method at 0, 60 and 120 cm from the row midline. Root density was greatest at a depth between 21 and 60 cm. The cumulative root fraction for root density registered a β value, a numerical quantity that summarises depth distribution, ranging between 0.932 and 0.962. Root number and density were significantly lower for the self-rooted vines compared to that of the grafted vines.

Conclusions: The scion genotypes affected most developmental parameters, including the diameter of the root system, the root density at 21–80 cm depth and the ratio of fine roots to coarse roots.

Significance of the Study: The scion plays an important role in grapevine root growth, development and distribution in a heavy clay soil, although the mechanism remains unclear.

Keywords: *cumulative root fraction, Nerello Mascalese, Nero d'Avola, root density,* Vitis vinifera

Introduction

A root system performs a multiplicity of functions—it anchors the plant allowing it to retain a fixed posture, absorbs water and minerals from the soil, synthesises organic compounds, including hormones which affect the growth of the shoot and serves as a store for reserve carbohydrates and mineral nutrients. Root system structure, function, mass, growth and spatial distribution all differ significantly between species and between cultivars and even between genetically identical individuals (Warschefsky et al. 2016). Root system structure especially influences the volume of the soil mass from which plants absorb water and minerals (Perkons et al. 2014) and, vice versa, the soil characteristics can affect the volume of soil available to the plant. Most of the root distribution is determined by soil conditions and depends on genetic differences in sensitivity to pH, oxygen, water, nitrogen and bulk density. The root system ensures the survival of the plant under a range of conditions of both the soil and the atmospheric environment (Saayman and Van Huyssteen 1982, Ball et al. 2005, Gregory 2006).

For perennial fruit crops, grafting a stock to a scion is a common means of overcoming limitations of the soil and atmospheric environments in relation to the biological 'fitness' of the scion genotype and also to obtain maximum agronomic and economic advantage from the chosen scion (Zohary and Spiegel-Roy 1975, Galet 1979). In modern viticulture, choice of an appropriate rootstock is key for vineyard efficiency because of its influence on the growth and properties of both rootstock and scion (Mudge et al. 2009) and appropriate resistance to environmental and biotic stresses. Usually in viticulture, a rootstock is selected for its rooting traits, for tolerance to soil pests and diseases (Warschefsky et al. 2016) or to limit the susceptibility of the vine to abiotic stresses such as drought or soil pH (Koepke and Dhingra 2013). Rootstock choice may also be based on its effects on ripening and on crop quality characteristics (Warschefsky et al. 2016).

The rooting density is influenced by both genetic factors (Southey 1992) and agronomic practices (Giulivo and Pitacco 1996). If planting density increases, there is a modification of the root's physical parameters and, in particular, root biomass because of the angle of root penetration and root length density (Archer and Strauss 1985, Morano and Kliewer 1994, Hunter 2000) and also of rooting depth (Southey and Archer 1988). If the soil characteristics are not particularly limiting, then the genotypic properties of the rootstock may predominate in determining root system development (Swanepoel and Southey 1989). A review of rooting depth in many root profiles of many rootstocks showed only limited genotype effects (Smart et al. 2006). Tandonnet et al. (2010) reported a large body of research on rootstock effects on scion vigour, yield components and physiological behaviour.

Some studies have highlighted the close interdependence of shoot growth and root growth, a relationship that differs between species, environment and plant age and size (Gedroc

doi: 10.1111/ajgw.12415
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et al. 1996, Hermans et al. 2006). A main effect is that any reduction in root system growth also reduces the growth of the shoot (Saayman and Van Huyssteen 1982, Richards 1983). Therefore, rootstock behaviour is related to its vigour and functionality (Jin et al. 2016).

Despite extensive studies of rootstock effect on scions, the effect of the scion on the rootstock has not been studied widely. This is largely because of the extreme difficulty associated with observing and accurately measuring root system growth and structure (Tandonnet et al. 2010). These authors showed, however, a strong effect of the scion on the biomass allocation between shoots and the root system, on leaves and the primary and lateral stem, and also on rootstock growth. This confirmed the previous observation of Oslobeanu (1978) who reported on the influence of the grafted scion cultivar on the rooting pattern in grapevine. Moreover, the scion appears to affect root growth and density at different soil depths (Swanepoel and Southey 1989), the zone of maximum root concentration, the size of the total root system (Daulta and Chauhan 1980) and the root necrosis of SO4 rootstock grown in trenches (Zapata et al. 2001).

For self-rooted vines, Southey and Archer (1988) reported high salt uptake, a low root number and deep root growth of the cultivar Moscato Rosso. This may be due to the low salt-exclusion capacity of self-rooted vines (Stevens et al. 1996). The low root system efficiency of self-rooted plants has also been reported for phosphorous and iron adsorption (Bavaresco and Lovisolo 2000).

Here we examine the root system of three rootstocks, 34 Ecole de Montpellier (34 E.M.), 140 Ruggeri (140 Ru.) and 1103 Paulsen (1103 P.), grafted to the scion cultivars Nerello Mascalese and Nero d'Avola, the most widely cultivated black grapes in Sicily, and the root systems of the same self-rooted cultivars. Vines were analysed using the profile wall method (Böhm 1979) involving multilayered excavations. The latter are rare in comparison to single wall profiles at a set distance from the trunk (Smart et al. 2006). The aims of this research were to evaluate the effect of the scion–rootstock interaction on: (i) the distribution of the root system in a heavy soil; (ii) the vegetative and productive performance of the scion; and (iii) the effect of the scion on the behaviour of the root system.

Materials and methods

Site, plant material and experimental design

Our investigations took place at the end of a long research period in the 17-year-old experimental vineyard at Catania University, located on the east coast of Sicily, South Italy (latitude 37°24′ 32.52″N; longitude 15°03′16.95″E; 0 masl). The climate is Mediterranean with mild and wet winters, while the summer is semi-arid with almost zero rainfall (Figure 1). The vineyard was planted with several local and international scion cultivars grafted onto 34 E.-M. (*Vitis berlandieri* Planch × *Vitis riparia* Michx), 140 Ru. and 1103P.(*V. berlandieri* Planch × *V. rupestris* Scheele) rootstocks. The rootstock 34 E.M. is not considered a highly vigorous rootstock (Carbonneau 1985), 140 Ru. is reported to have a good adaptability to conditions of high drought level and chlorosis (Chauvet and Reynier 1979, Carbonneau 1985), while 1103P. is well adapted to drought and salt stress (Lavrenčič et al. 2007). The scion cultivars examined were Nerello Mascalese and Nero d'Avola, both grafted to the above rootstocks and self-rooted. Both cultivars are mid-late ripening and highly vigorous, but Nero d'Avola is better adapted to warm and drought climate areas (Nicolosi et al. 2012).

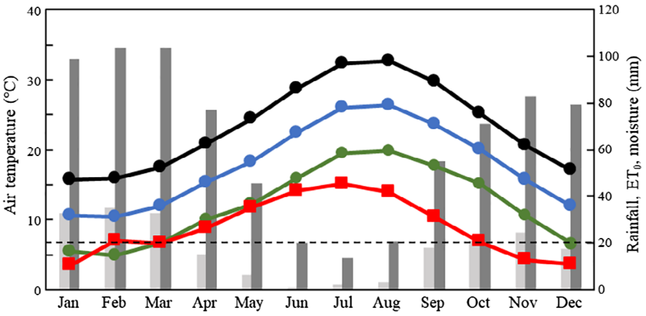

Figure 1. Monthly minimum (●), mean (●) and maximum (●) air temperature, rainfall (■), reference evapotranspiration (ET$_o$) (■) and moisture (■) registered at the experimental vineyard of the University of Catania (latitude 37° 24′32.52″N; longitude 15°03′16.95″E; masl 0). The crop is considered to be water stressed below 20% maximum plant available water.

The vines were planted at 2.5 m spacing between rows and 1.2 m between vines within rows, giving 3333 vines/ha. Rows were oriented north–south and trained to a unilateral cordon at 0.8 m with a canopy height of approximately 1.9 m. Vines were spur-pruned to eight to ten nodes per vine (two nodes per spur and four to five spurs). The shoots were vertically positioned.

All agronomic practices were applied uniformly and in accordance with standard cultural practices in the area. Soil management comprised mechanical tilling (15 cm) two or three times each year with periodic weed control in the row using a string-trimmer. Sprinkler irrigation was applied during the dry period in mid-summer. Vines received irrigation as needed to maintain soil moisture adequate for canopy growth and full ripening of the crop. Based on climate conditions, field observations and on experience, a maximum of three irrigations between July and August was applied comprising 30–40 L per vine for each irrigation. Four vines per rootstock and/or self-rooted plants were arranged in a completely random design.

Soil analysis

Soil characteristics were measured at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm), with three replications. Organic matter (OM) was measured by quantifying total organic carbon (mg/kg) according to Springer and Klee (1954). Soil extractable phosphorus (mg/kg) was determined according to Olsen et al. (1954). Soil exchangeable potassium (meq/100 g) was determined in a solution of barium chloride and triethanolamine at pH 8.2 (2 g of soil: 25 mL). After the total calcium carbonate ($CaCO_3$) was measured by the gas-volumetric method using a Dietrich–Fruhling calcimetre, the active $CaCO_3$ (%) was quantified using a 0.2 N ammonium oxalate solution (Loeppert and Suarez 1996). The available soil moisture (mm) was calculated as the difference between the field capacity and the wilting point in the soil bulk density obtained at each of the five layers (Scordia et al. 2017). The pH and electrical conductivity (EC), measured on a soil aqueous extract, were determined according to European standards 13 037 (European Committee for Standardization 2012a) and 13 038 (European Committee for Standardization 2012b), respectively, using an HI

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9813 portable EC meter (Hanna Instruments, Woonsocket, RI, USA) and an AB 15 pH meter (Thermo Fisher Scientific, Waltham, MA, USA).

Vine monitoring

Destructive measurements were made of vine growth on 20 vines per treatment at the end of flowering (E-L 26) (Coombe 1995), at late spring. The trunk circumference of the rootstock and scion was measured about 5 cm above and just below the graft union. The number, the mass and the basal diameter of main shoots and the total above-ground vine (trunk, branches and 1-year-old wood) mass were recorded. Once the vegetative material was excised, fresh mass was measured immediately.

Root system investigations

Root systems were studied according to the method described by Böhm (1979). Four vines with similar trunk diameter were selected for each treatment (cultivar, rootstock, self-rooted) just after the above measurements were recorded (E-L stage 26). Two trenches were dug with an excavator, one on each side of the row, to a depth of 1.50 m and a length of 1.40 m. After observations were made of wall sections 1 m deep and 1.20 m long on both sides of the row a full excavation was completed. Soil was removed manually with high pressure (300 kPa) water sprays. Observations were made at a distance of 120 cm (profile 120), at 60 cm (profile 60) and at 0 cm (profile 0) from the centre of the row, that is in the plane of the row axis. After soil removal for each profile, a sub-quadrat grid (20 × 120 cm, i.e. 0.24 m^2) was fixed in place and the roots were counted and their positions recorded within the grid. For the 120 cm profile exposed roots were traced to exclude those roots from the vines in the adjacent row. For fine roots (<2 mm), only those belonging to the main roots (>2 mm) were counted.

Roots were categorised after Morlat and Jacquet (1993) with some modifications using two diameter classes: 0.5–2 mm and >2 mm. Root density was calculated and expressed as number/m^2 (Southey 1992, Morlat and Jacquet 1993) of trench wall surface and referenced with respect to five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm). For each wall profile (120, 60 and 0), in order to evaluate the relative ratio of fine to larger roots of the root systems, the rooting index (RI) was calculated as the ratio of the number of roots <2 mm/number of roots >2 mm (Swanepoel and Southey 1989). A high RI indicates positive soil conditions and a better utilisation of the available soil. After the root distribution measurements were completed, the total root system was weighed and the ratio of the root system mass/vine above-ground mass and the ratio of the root system mass/shoot mass were recorded. Values were expressed as fresh mass. Root systems were also observed for phylloxera symptoms.

Long-term scion performance

Over the 10-year period prior to the destructive measurements described above, the yield parameters and compositional measures of grape bunches had been recorded at ripening (mid-September). All bunches per vine were counted and weighed and the yield/vine calculated. The winter pruning mass material was registered. Fruit composition parameters were determined: TSS with a digital refractometer with temperature correction (model RX-5000 Atago, Tokyo, Japan), pH and TA, expressed as g/L of tartaric acid equivalents, with an automatic titrator (Titrino model 798, Metrohm, Riverview, FL, USA) with a 5.0 mL juice sample being titrated against 0.1 mol NaOH to pH 8.2.

Statistical analysis

The significance of each variable ($P \leq 0.05$) was tested by ANOVA with StatSoft 6.0 (StatSoft Italia2005, Vigonza, Padova, Italy). Separation of means was calculated using Tukey's honest significant difference (HSD) test. Significant effects on root number and RI, vine vegetative behaviour, vine performance and yield components were calculated by a factorial ANOVA. For the factorial ANOVA two or more independent variables were tested for possible interaction effects on a single dependent variable. For below-ground measures, a factorial ANOVA with four independent variables—(i) Cultivar; (ii) Rootstock; (iii) Profile; (iv) Depth—and the interactions between them was conducted.

The cumulative fraction of roots at increasing depth was calculated by a model equation previously used to describe the vertical root distributions of trees (Gale and Grigal 1986) and, more recently, of grapevines (Smart et al. 2006).

The cumulative fraction of roots as a function of soil depth was tested by its fit to the model $Y = (1 - \beta^d)$, where Y is the cumulative fraction of roots with depth and d is soil depth in cm. The estimated coefficient β can be used as a numerical quantity that summarises depth distribution, where higher values for β correspond with a greater proportion of roots at depth (Smart et al. 2006). The equation $Y = (1 - \beta^d)$ was estimated for each of the root systems studied. For β determination the assumption of normality was checked with the Kolmogorov–Smirnov's test. Significant results for the variables tested suggested that hypothesis had been violated. Therefore the data were square root transformed to satisfy the assumption of normality.

Results

Soil characteristics

The vineyard soil analysis (Table 1) according to the United States Department of Agriculture (USDA) scheme (United States Department of Agriculture 1975), showed the soil profile composition as: 0–20 cm (clay loam), 21–40 cm (clay loam), 41–60 cm (clay), 61–80 cm (clay loam) and 81–100 cm (clay). The highest skeleton (stones >2 mm) content was found between 41 and 80 cm depth. The drainage category was 'somewhat poorly drained'. The bulk density was less than 1 g/cm^3 at 0–20 depth, while it did not differ significantly between the lower four depths (21–40, 41–60, 61–80 and 81–100 cm) where it was close to 1.50 g/cm^3. The organic matter was highest at 3.1% in the 41–60 cm layer. The pH was moderately alkaline in the top two layers and weakly alkaline below this. The $CaCO_3$ content and EC were high, especially in the deepest layer 81–100 cm. No evidence of phylloxera was observed in any soil layer and no nodes were observed on the roots of the self-rooted vines.

Root density

The overall pattern of root density with soil depth was similar in all scions and rootstocks with the greatest number of roots generally in the 40–60 cm layer, where OM content was highest (Table 1). In the upper soil layer (0–20 cm) in

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Table 1. Effect of depth on the main physical and chemical characteristics of the soil at Catania University experimental vineyard, Catania, Italy.

Depth (cm)	Skeleton (>2 mm) (g/kg)	Fine particles (<2 mm) (g/kg)	Clay (%)	Silt (%)	Sand (%)	Bulk density (g/cm^3)	Soil moisture content (mm)	Organic matter (%)	pH	Active carbonate ($CaCO_3$) (%)	Electrical conductivity 25°C (dS/m)	Exchangeable potassium (meq/100 g)	Extractable phosphorous (mg/kg)
0–20	5	995	31.27	30.25	38.48	0.98	8.6	1.6	7.88	2.4	0.65	0.4	542
21–40	7	993	39.37	29.61	31.02	1.51	15.9	2.2	7.93	1.7	0.71	0.4	553
41–60	10	990	51.61	25.36	23.03	1.46	16.5	3.1	7.74	2.5	0.82	0.2	452
61–80	13	987	33.53	35.27	31.20	1.54	13.5	2.6	7.86	3.0	0.82	0.2	325
81–100	3	997	53.26	23.87	22.87	1.46	15.3	1.8	7.47	3.2	1.83	0.2	356

the wall profiles of 0 and 120 cm from the centre of the row, the 34 E.M. rootstock had the highest root density for both Nerello Mascalese and Nero d'Avola (Figure 2). The self-rooted vines of both cultivars showed the lowest root density at all depths. In the wall profile 60 cm from the row axis, the highest root density was for the 1103 P. rootstock in combination with Nerello Mascalese in the layers 21 and 60 cm and 81–100 cm. In the wall profile 0 (nearest to the main stems), the 1103 P. rootstock had the highest root density with both scion cultivars at all depths except the shallowest (0–20 cm). Except in the 0–20 cm layer, the self-rooted vines of both cultivars had the lowest root density.

The estimated β value (higher values correspond to a greater proportion of roots at depth) (Figures 3, 4) for the cumulative fraction of root density among rootstocks, self-rooted vines, cultivars and profiles ranged between 0.952 and 0.958 for Nerello Mascalese and between 0.955 and 0.962 for Nero d'Avola (Table 2). At the wall profile 120 both for Nerello Mascalese and Nero d'Avola the β value was similar for each root system. At the wall profile 60 between the two cultivars the best cumulative root fraction was reached by the self-rooted vines of Nero d'Avola. At the wall profile 0, the cumulative root fractions of 34 E.M., 1103 P. and 140 Ru. with Nerello Mascalese were similar to those at 120. The 140 Ru. rootstock grafted with Nerello Mascalese showed the highest total root number value for all wall profiles (Figures 5, 6), while the self-rooted vines had the lowest values. In wall profiles 120 and 60, the 1103 P. and 34 E.M. rootstocks differed significantly from 140 Ru. and the self-rooted vines. The 34 E.M. and 140 Ru. rootstocks grafted with Nero d'Avola had the highest root numbers at wall profile 0.

Overall, Nero d'Avola had a higher RI than Nerello Mascalese (Figures 5, 6) because of more fine roots rather than fewer larger roots. Nero d'Avola grafted on 34 E.M. showed the highest RI (number of roots <2 mm/number of roots >2 mm) in each profile indicating a relatively finer root system. Nerello Mascalese 34 E.M. showed relatively low RI. At the wall profiles 120 and 0, the highest RI was recorded for Nerello Mascalese grafted on 1103 P. and for the self-rooted vines. The 34 E.M. and 140 Ru. rootstocks at wall profile 60 had the highest RI. Nero d'Avola grafted on 34 E.M. showed the highest RI in each profile.

Vegetative and reproductive growth

The self-rooted Nerello Mascalese and Nero d'Avola vines had the lowest trunk circumference for both scion and rootstock (Table 3). These vines, however, also possessed the highest number of main shoots. The diameter of the shoots decreased significantly for the self-rooted vines of Nero d'Avola. The above-ground mass of all grafted Nerello Mascalese vines was significantly higher than that of the self-rooted vines. Only Nero d'Avola on 34 E.M. exhibited higher above-ground mass compared to that of the self-rooted vines. The total root system mass was significantly higher for 140 Ru. and 1103 P. in combination with both Nerello Mascalese and Nero d'Avola. The highest root system mass/shoot mass ratio was observed on 34 E.M. and 140 Ru. with Nerello Mascalese and on 1103 P. for Nero d'Avola (Table 3).

Mean harvest yield was highest for Nerello Mascalese on 140 Ru. The self-rooted Nerello Mascalese vines and

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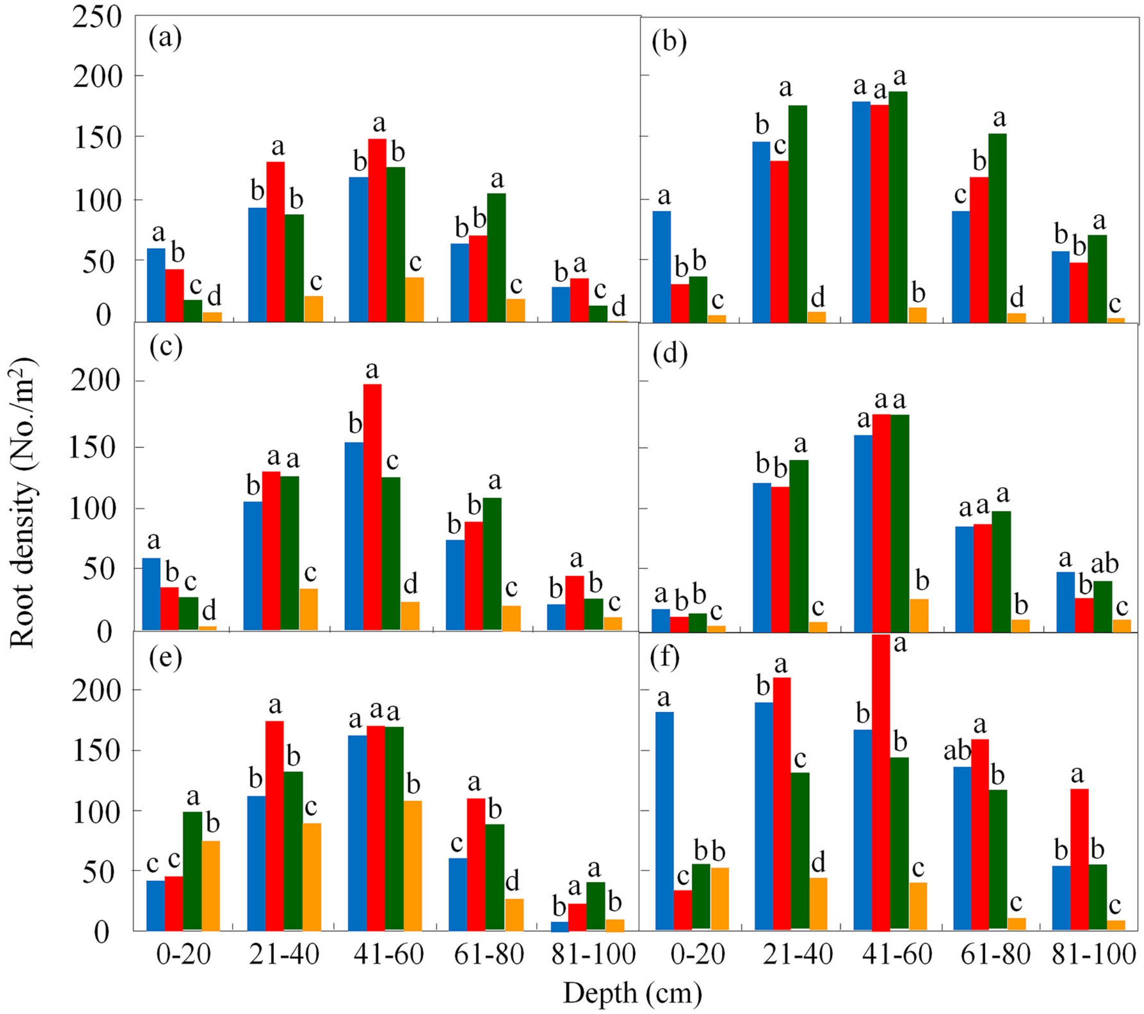

Figure 2. Effect of cultivar, root system, soil depth and distance from the mid-row on root density of the trench wall surface after a sub-quadrat grid (20 × 120 cm, i.e. 0.24 m^2) was fixed in place. The scion cultivars, (a, c, e) Nerello Mascalese and (b, d, f) Nero d'Avola, were self-rooted (🟨) and grafted onto the rootstocks, 34 Ecole de Montpellier (*Vitis berlandieri* Planch × *Vitis riparia* Michx) (🟦), 140 Ruggeri (🟩) and 1103 Paulsen (🟥). Measurements were made at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm) and at three distances, (a,b) 120, (c,d) 60 and (e,f) 0 cm, from the row midline. Different letters on the bars at each soil depth indicate significantly different mean values of root density among grafted and self-rooted vines based on Tukey's honest significant difference test ($P \leq 0.05$).

both the self-rooted and 1103 P. grafted Nero d'Avola vines had the lowest yield. The yield/main shoot ratio was higher for 34 E.M. grafted to Nerello Mascalese and 140 Ru. vines grafted to Nero d'Avola. The TSS was lowest for Nerello Mascalese on 34 E.M. and Nero d'Avola on 140 Ru., while Nerello Mascalese on 140 Ru., 1103 P. and self-rooted Nero d'Avola recorded the lowest TA (Table 4).

Discussion

The physicochemical analysis of the soil (Table 1) confirmed that it was a heavy textured clay, with high bulk density and high $CaCO_3$ content (not shown). As reported elsewhere, root distribution depends on soil water and nutrient availability (Bengough et al. 2006), while root elongation is influenced by soil moisture (Morlat and Jacquet 1993). With a clay content of 53.26% at 81–100 cm depth, the heavy texture and bulk density likely reduced root system density due to hypoxic conditions (Bengough et al. 2006) and after drying hardness probably reduced root growth. The active $CaCO_3$ content and the EC from the 0–80 cm depth were not likely to limit root development and crop yield. At the deepest layers, however, from 81 to 100 cm, these levels were likely to restrict growth (Maas and Hoffman 1977) and especially for the self-rooted vines. The active $CaCO_3$ in the 80–100 layer was similar to that of the 61–80 cm layer. Hence, the role played by EC as a limiting factor was likely to be greater. In general the OM content between 21 and 80 cm depth had a mean value of 2.6% and probably had a greater influence on roots in these layers.

For self-rooted vines the results agree with those of Prior et al. (1992) who showed that in heavier soils, high soil moisture can be a significant limitation in comparison to rootstock root systems (Figures S1,S2). In particular, the high EC and the active $CaCO_3$ negatively influenced vine growth in that study. Under the conditions described, the 140 Ru. and the 1103 P. rootstocks showed greater growth in the deeper layers, which is consistent with their apparent high tolerance of active $CaCO_3$ (Ksouri et al. 2005) and salt (Walker et al. 2002). Also the 34 E.M. rootstock demonstrates a low drought tolerance (Carbonneau 1985), but is adaptable to a wide range of soils. Conversely, the low root density in the

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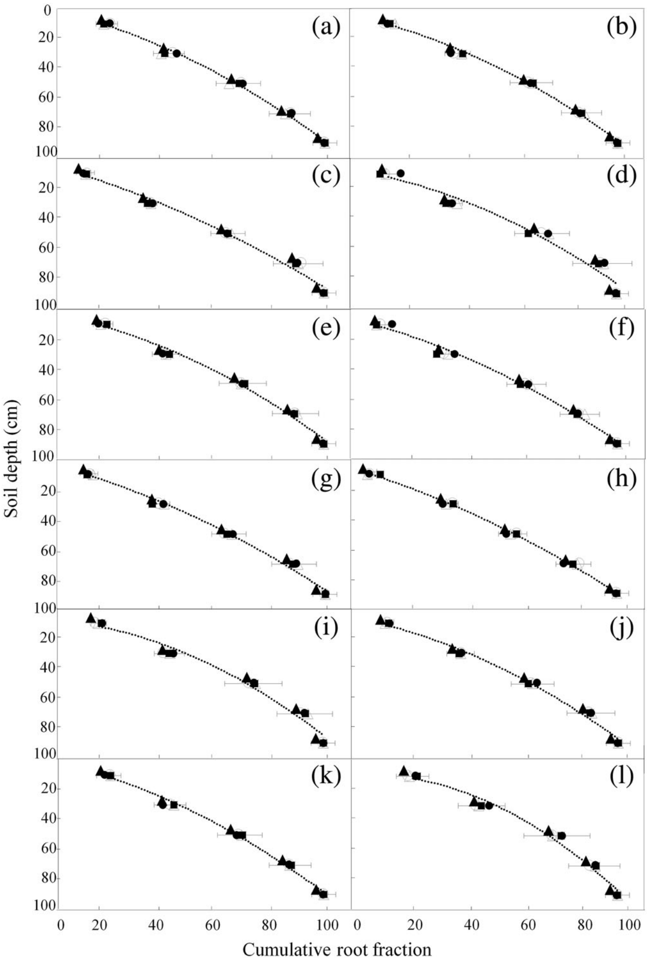

Figure 3. Effect of root system, soil depth and distance from the mid-row on the cumulative root fraction of the total root density of the cultivar Nerello Mascalese. The root system was (d, h, l) self-rooted and the three rootstocks (a, e, i) 34 Ecole de Montpellier (*Vitis berlandieri* Planch × *Vitis riparia* Michx), (c, g, k) 140 Ruggeri and (b, f, j) 1103 Paulsen. Measurements were made at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm) and at three distances: (a, b, c, d) 120, (e, f, g, h) 60 and (i, j, k, l) 0 cm, from the row midline. The data are fitted to the distribution model $Y = (1 - \beta^d)$ of Gale and Grigal (1986), where Y is the cumulative fraction of roots with depth and d is the soil depth (cm). Bars represent SD of the replicates 1 (○), 2 (Δ), 3 (■), 4 (●) and mean (▲). R^2 values are: (a) 0.9958; (b) 0.9958; (c) 0.9848; (d) 0.9678; (e) 0.9899; (f) 0.994; (g) 0.9927; (h) 0.9991; (i) 0.9779; (j) 0.9911; (k) 0.9953; and (l) 0.9855.

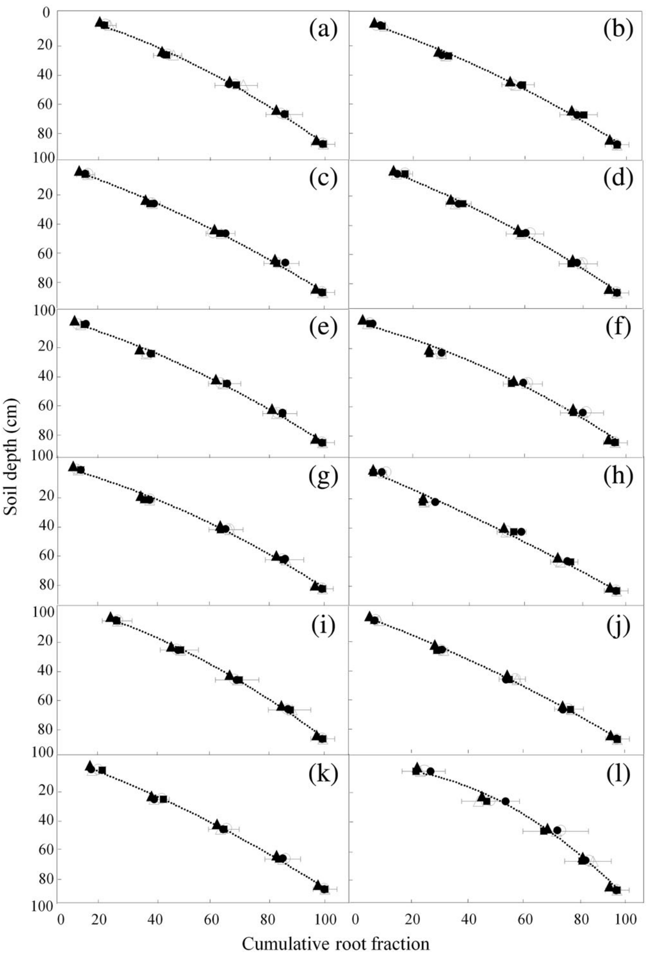

Figure 4. Effect of root system, soil depth and distance from the mid-row on the cumulative root fraction of the total root density of the cultivar Nero d'Avola. The root system was (d, h, l) self-rooted and the three rootstocks (a, e, i) 34 Ecole de Montpellier (*Vitis berlandieri* Planch × *Vitis riparia* Michx), (c, g, k) 140 Ruggeri and (b, f, j) 1103 Paulsen. Measurements were made at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm) and at three distances, (a, b, c, d) 120, (e, f, g, h) 60 and (i, j, k, l) 0 cm, from the row midline. The data are fitted to the distribution model $Y = (1 - \beta^d)$ of Gale and Grigal (1987), where Y is the cumulative fraction of roots with depth and d is the soil depth (cm). Bars represent SD of the replicates 1 (○), 2 (Δ), 3 (■), 4 (●) and mean (▲). R^2 values are: (a) 0.9958; (b) 0.9958; (c) 0.9848; (d) 0.9678; (e) 0.9899; (f) 0.994; (g) 0.9927; (h) 0.9991; (i) 0.9779; (j) 0.9911; (k) 0.9953; and (l) 0.9855.

upper soil layers (0–20 cm) was probably because of mechanical tillage (15 cm deep) which destroys the superficial roots and the associated increase in soil drying and high temperature in the shallow layers. Such responses were less obvious for 34 E.M. than for Nero d'Avola at this depth (0–20). The wall profile 0 appeared to have good root regeneration after tillage considering that the last tillage was made in early spring (end of March). Both root number and root size were elevated in the mid-levels (21–80 cm depth) for all rootstocks (140 Ru., 1103 P. and 34 E.M.) compared with that of the self-rooted vines. The high root density at the mid layers was well correlated with the high level of OM registered in the soil. Based on the values reported in Figure 2, the soil appeared to influence root density and distribution, mainly in the deeper layers.

In this study the constant spacing, mature age and history of cropping of the vine represented only three factors for predicting soil colonisation. The trunk diameter indicated that the vines were small in respect to the vine age (17 years). Therefore, the root development and distribution between the different profiles and depths was because of the interaction among the edaphic traits, the cultural practices and the scion/rootstock genetic components.

The vertical root development of both cultivars and of self-rooted vines had a good distribution over the top metre of soil that was excavated as revealed by the fitted β values and regression models (Figures 3, 4). A similar distribution was found by Smart et al. (2006) even when the excavation depth was greater. Thus, it is possible that some roots can colonise deeper layers. The β values were close to 1 and this corresponded to a higher proportion of root growth at the deeper layers. In particular, the observed β values were always higher than 0.92 and less than 0.97 and associated with a larger proportion of roots at the soil surface zone and at the deeper layers (Gale and Grigal 1986). In this study generally 90% of the observed roots were in the top

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Table 2. Cumulative root fraction of the root density as a function of soil depth for each cultivar, profile and root system.

Profile	Root system	Cumulative root fraction (cm) 0–20	21–40	41–60	61–80	81–100	β†
Nerello Mascalese							
120	34 Ecole de Montpellier	16.5 ± 3.7	42.2 ± 7.1	74.2 ± 11.4	92.2 ± 9.9	100.0 ± 4.5	0.955 ± 0.003abc
	140 Ruggeri	5.3 ± 2.2	30.3 ± 1.9	66.5 ± 7.7	96.1 ± 11.7	100.0 ± 4.5	0.956 ± 0.001abc
	1103 Paulsen	10.3 ± 1.9	40.2 ± 5.8	75.5 ± 11.4	91.7 ± 9.7	100.0 ± 4.5	0.955 ± 0.001abc
	Self-rooted	8.9 ± 3.7	32.1 ± 2.4	78.1 ± 13.5	98.9 ± 12.9	100.0 ± 4.5	0.955 ± 0.002abc
	P-value						***
60	34 Ecole de Montpellier	14.3 ± 3.0	39.8 ± 4.7	77.1 ± 12.2	94.9 ± 11.1	100.0 ± 4.5	0.958 ± 0.001abc
	140 Ruggeri	6.4 ± 1.8	37.0 ± 4.7	67.5 ± 8.1	93.8 ± 10.7	100.0 ± 4.5	0.958 ± 0.001abc
	1103 Paulsen	6.8 ± 3.5	32.8 ± 4.3	73.3 ± 10.7	91.1 ± 9.6	100.0 ± 4.5	0.958 ± 0.001abc
	Self-rooted	3.4 ± 3.4	40.1 ± 5.2	65.4 ± 7.3	88.2 ± 8.9	100.0 ± 4.5	0.958 ± 0.001abc
	P-value						***
0	34 Ecole de Montpellier	10.9 ± 1.6	40.0 ± 5.0	82.2 ± 14.4	97.9 ± 12.5	100.0 ± 4.5	0.952 ± 0.002c
	140 Ruggeri	18.7 ± 4.2	43.6 ± 7.0	75.8 ± 11.6	92.4 ± 10.0	100.0 ± 4.5	0.952 ± 0.002c
	1103 Paulsen	8.8 ± 1.1	41.8 ± 5.4	74.2 ± 11.0	95.4 ± 11.3	100.0 ± 4.5	0.953 ± 0.001c
	Self-rooted	24.2 ± 6.8	53.2 ± 10.9	88.0 ± 17.1	96.7 ± 11.9	100.0 ± 4.5	0.954 ± 0.001cb
	P-value						***
Nero d'Avola							
120	34 Ecole de Montpellier	16.1 ± 3.1	42.1 ± 5.9	73.9 ± 11.2	89.8 ± 8.9	100.0 ± 4.5	0.958 ± 0.001abc
	140 Ruggeri	5.9 ± 2.0	34.1 ± 2.2	64.3 ± 3.2	88.8 ± 8.7	100.0 ± 4.5	0.958 ± 0.001abc
	1103 Paulsen	6.3 ± 2.0	32.1 ± 2.0	67.0 ± 8.0	90.2 ± 9.2	100.0 ± 4.5	0.958 ± 0.001abc
	Self-rooted	17.8 ± 4.5	40.2 ± 5.3	70.1 ± 9.5	89.6 ± 9.2	100.0 ± 4.5	0.958 ± 0.001abc
	P-value						***
60	34 Ecole de Montpellier.	4.3 ± 2.7	32.2 ± 1.4	69.0 ± 8.6	88.7 ± 8.5	100.0 ± 4.5	0.961 ± 0.001ab
	140 Ruggeri	3.2 ± 3.1	32.8 ± 2.7	70.3 ± 9.6	91.3 ± 9.5	100.0 ± 4.5	0.956 ± 0.002abc
	1103 Paulsen	3.0 ± 3.2	30.8 ± 3.4	72.4 ± 10.8	93.3 ± 10.6	100.0 ± 4.5	0.959 ± 0.004abc
	Self-rooted	6.6 ± 2.6	20.3 ± 5.1	65.5 ± 8.2	82.8 ± 6.8	100.0 ± 4.5	0.962 ± 0.001a
	P-value						***
0	34 Ecole de Montpellier	24.9 ± 6.7	51.0 ± 9.4	73.9 ± 10.7	92.6 ± 10.2	100.0 ± 4.5	0.956 ± 0.002abc
	140 Ruggeri	10.8 ± 2.6	37.1 ± 4.2	65.8 ± 7.2	89.1 ± 8.7	100.0 ± 4.5	0.955 ± 0.001abc
	1103 Paulsen	4.3 ± 2.6	31.7 ± 1.9	64.1 ± 7.0	84.7 ± 6.9	100.0 ± 4.5	0.956 ± 0.001abc
	Self-rooted	33.6 ± 10	61.6 ± 16.1	87.9 ± 17.2	94.7 ± 11.2	100.0 ± 4.5	0.956 ± 0.000abc
	P-value						***

†Beta values are calculated according the model of Gale and Grigal (1986): $Y = (1 - \beta^d)$, where Y is cumulative fraction of roots with depth and d is the soil depth (cm). Calculated β values ± SD indicated by different lower case letters are significantly different ($P \le 0.001$) based on Tukey's HSD test within cultivars, profile and rootstock/root.

21–80 cm and in particular at 41–60 cm depth. At 0–20 cm depth the low number of roots may have been because of the soil mechanical tillage and to the low soil moisture content, while in the deepest layers the EC appeared to be the highest limiting factor for root growth. Arbabzadeh and Dutt (1987) showed that the reduction in vine growth was about 8.4% for each 1.0 dS/m increase in salt concentration above a threshold value of 1.1 dS/m; therefore, the EC level measured on the soil aqueous extract of 1.83 dS/m likely reduced root growth.

A large collection of root maps of vertical trench walls was analysed by Smart et al. (2006) and generally the β value was above 0.98, as in most cases 90% of the roots were found in the top 120 cm. For 1103 P. and 140 Ru. Southey (1992) found a β of 0.98 in a sandy clay loam soil, while Southey and Archer (1988) found a value of 0.98 and 0.99, respectively, for 1103 P. and 140 Ru. in brown apedal and sandy loam soils. In a silt loam soil the value for 1103 P. was 0.98 (Swanepoel and Southey 1989). For the performance of the *Vitis vinifera* root system Padgett-Johnson (1999) found a β value of 0.98 in a sandy loam soil for the cultivar Carignan. No previous studies have been reported for the 34 E.M. rootstock.

In general, the three rootstocks did not differ in total roots while the self-rooted had fewer total roots. Among the profiles and cultivars, the highest root number was observed in the profiles 120 and 0 for Nero d'Avola grafted on 1103 P. or 34 E.M. rootstocks. Moreover Nero d'Avola exhibited a lower RI than Nerello Mascalese (Figures 5, 6), indicating a relatively finer root system regardless of rootstock due to more fine roots, not fewer larger roots. This may have been a result of the lower crop level in Nero d'Avola in all combinations compared to the higher crop level in Nerello Mascalese (Table 4). The higher pruning mass of Nero d'Avola similarly suggests a reduction in crop level on vegetative growth. As far as the horizontal development of root systems of the adjacent vines in different rows and in the same row, different responses were observed. In general, except in rare cases, no principal roots were traced from the adjacent row. Nevertheless, when the profile 120 was assessed, despite attempts to follow the main roots until the profile 0, we could not exclude the contemporary presence of root systems from the vines in the adjacent row. In contrast strong root systems were observed for the vines next to the studied ones. In these cases the foreign roots were painted white (Figures S1,S2).

Taken together, our results suggest a higher root systems distribution of all the graft combinations in the deeper and heavier soil layers and, compared to self-rooted, suggesting greater absorption of water and nutrients in the deepest zones (Yue et al. 2006). Moreover these results suggest an influence of the scion on rooting pattern due to an high growth and yield/vine (Tandonnet et al. 2010) that is regulated by a relationship between root and shoot activity (Wilson 1988) (Tables S1–S3). For rootstocks grafted with

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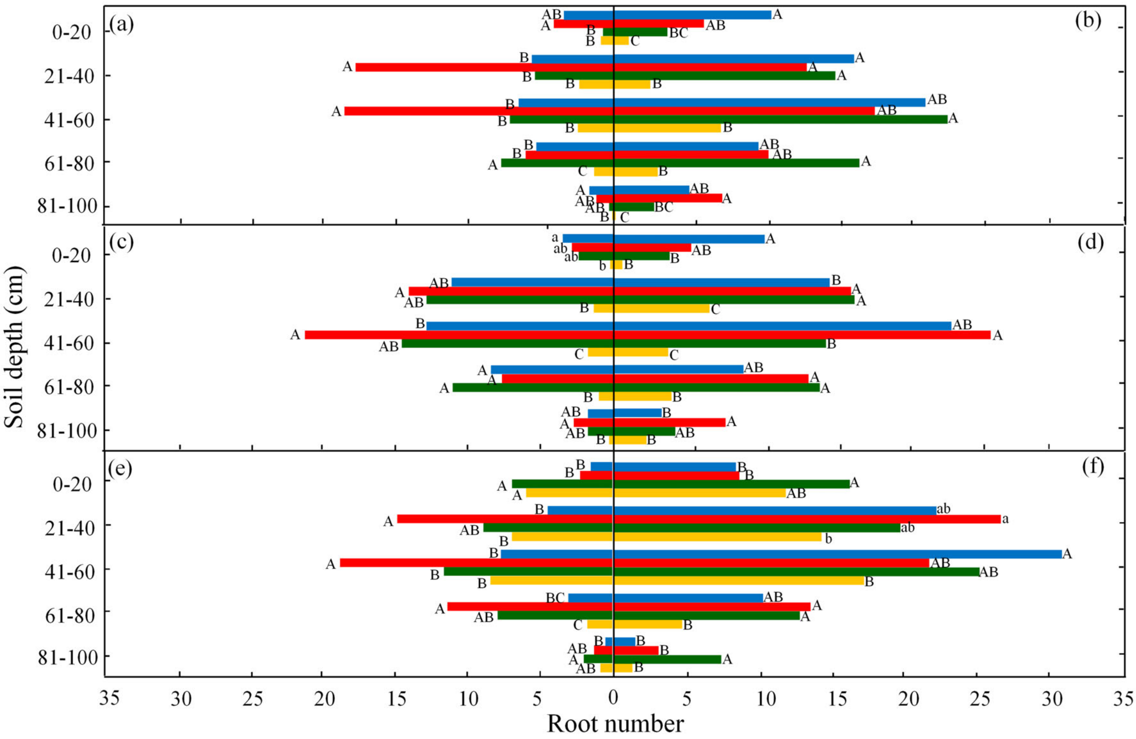

Figure 5. Root number observed for each sampling area for the scion cultivar Nerello Mascalese within two diameter classes [(a, c, e) <2 mm or (b, d, f) >2 mm], at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm) and three distances from the row midline [wall profiles (a,b) 120, (c,d) 60 and (e,f) 0 cm from the midline]. Nerello Mascalese was on self-rooted (🟨) and grafted onto the rootstocks, 34 Ecole de Montpellier (*Vitis berlandieri* Planch × *Vitis riparia* Michx) (🟦), 140 Ruggeri (🟩) and 1103 Paulsen (🟥). Mean values for each histogram with different letters are significantly different at $P \leq 0.05$ (lowercase letters) and at $P \leq 0.001$ (capital letters) level based on Tukey's honest significant difference test within a cultivar profile and depth.

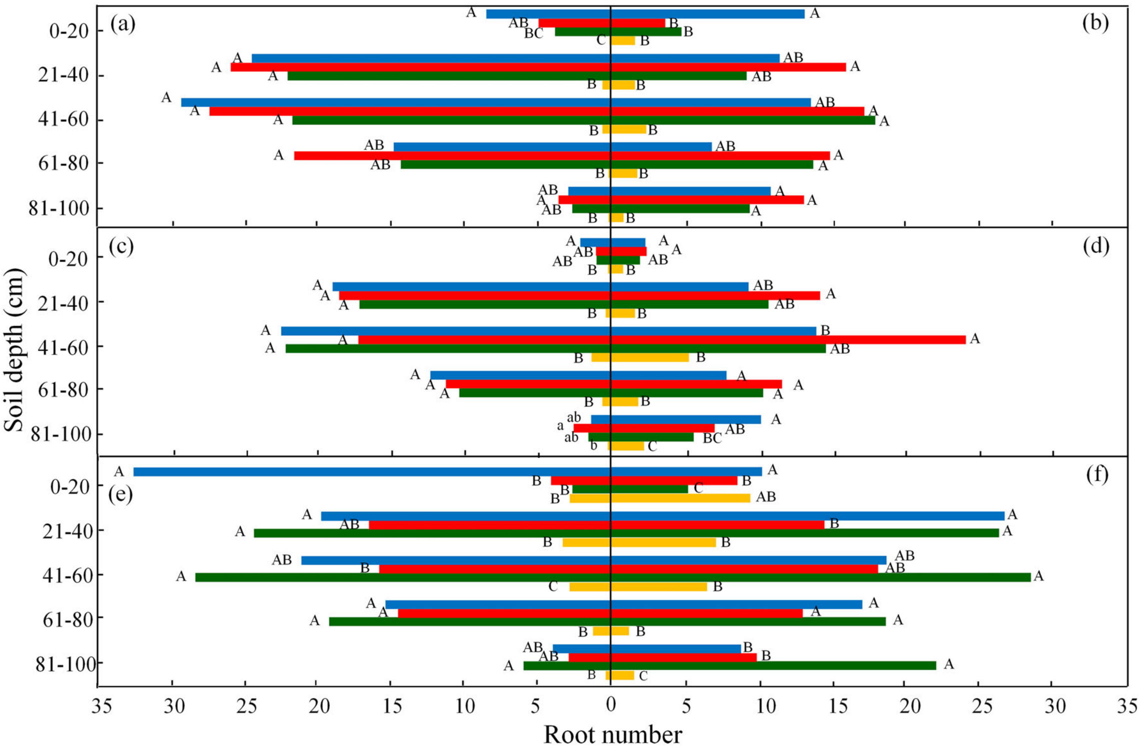

Figure 6. Root number observed for sampling area for the scion cultivar Nero d'Avola within two diameter classes [(a, c, e) <2 mm or (b, d, f) >2 mm], at five depths (0–20, 21–40, 41–60, 61–80 and 81–100 cm) and three distances from the row midline [(wall profiles (a,b) 120, (c,d) 60 and (e,f) 0 cm from the row midline]. Nero d'Avola was on self-rooted (🟨) and grafted onto the rootstocks 34 Ecole de Montpellier (*Vitis berlandieri* Planch × *Vitis riparia* Michx) (🟦), 140 Ruggeri (🟩) and 1103 Paulsen (🟥). Mean values for each histogram with different letters are significantly different at $P \leq 0.05$ (lowercase letters) and at $P \leq 0.001$ (capital letters) level based on Tukey's honest significant difference test within a cultivar profile and depth.

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Table 3. Effect of root system on the main vegetative behaviour of Nerello Mascalese and Nero d'Avola†.

Cultivar/root system	Rootstock trunk circumference (cm)	Scion trunk circumference (cm)	Main shoots (No.)	Shoot mass (kg)	Shoot basal diameter (mm)	Vine above-ground mass (kg)	Root system mass (kg)	Root system mass/vine above-ground mass	Root system mass/ shoot mass
Nerello Mascalese									
34 Ecole de Montpellier	19.3 ± 3.2AB	17.6 ± 2.5AB	6.5 ± 1.1ABC	0.12 ± 0.1D	9.7 ± 0.5ABC	3.42 ± 0.52a	2.11 ± 0.32C	0.6 ± 0.19	17.1 ± 2.3a
140 Ruggeri	21.0 ± 2.4A	17.3 ± 3.4ABC	6.5 ± 0.9 BC	0.26 ± 0.1 BC	8.6 ± 0.7C	3.50 ± 0.24a	3.81 ± 0.21A	1.1 ± 0.12	14.9 ± 1.9a
1103 Paulsen	19.8 ± 4.3A	20.6 ± 3.2A	7.5 ± 1.3ABC	0.29 ± 0.1ABC	8.9 ± 1.0 BC	3.19 ± 0.68a	3.79 ± 0.29A	1.2 ± 0.21	13.2 ± 2.6ab
Self-rooted	15.7 ± 2.4C	16.3 ± 2.4 BC	8.0 ± 1.2A	0.18 ± 0.1CD	8.8 ± 0.9 BC	2.91 ± 0.21b	2.35 ± 0.33 BC	0.8 ± 0.15	13.0 ± 2.4ab
Nero d'Avola									
34 Ecole de Montpellier	16.0 ± 3.1 BC	17.3 ± 4.2ABC	7.5 ± 1.0ABC	0.38 ± 0.1AB	10.1 ± 1.2AB	4.45 ± 0.31a	2.76 ± 0.65 BC	0.62 ± 0.82	7.2 ± 1.5c
140 Ruggeri	19.5 ± 1.4AB	19.2 ± 3.2AB	7.5 ± 1.2AB	0.39 ± 0.1A	10.7 ± 1.3A	3.73 ± 0.65ab	3.03 ± 0.35AB	0.81 ± 0.36	7.9 ± 1.3c
1103 Paulsen	16.0 ± 2.3 BC	16.0 ± 3.6 BC	6.0 ± 1.4 BC	0.33 ± 0.1AB	10.5 ± 1.0A	3.90 ± 0.42ab	3.15 ± 0.14AB	0.81 ± 0.20	10.0 ± 1.6bc
Self-rooted	14.1 ± 2.6C	13.2 ± 3.1C	8.0 ± 1.0AB	0.30 ± 0.1ABC	8.7 ± 0.8C	2.86 ± 0.19b	2.08 ± 0.19C	0.73 ± 0.46	7.1 ± 1.2c
n	20	20	20	20	4	4	4	4	4
P-value	***	***	***	***	***	*	**	ns	*

†Values are means ± SD of measurements made at the end of flowering (E-L stage 25). Mean values in the same column indicated by different letters are significantly different at $P \leq 0.05$ (*, lowercase letters), at $P \leq 0.01$ (**,uppercase letters) and $P \leq 0.001$ (***, uppercase letters) level; in absence of letters there are no significant differences (ns) among the means based on Tukey's honest significant difference test for factorial ANOVA.

Nerello Mascalese we found larger roots in the shallower depths than for Nero d'Avola. This suggests that Nero d'Avola has a better adaptation to heavy soils when grafted on the three rootstocks studied compared to that of self-rooted vines.

The higher growth of the 140 Ru. and 1103 P. rootstocks as seen in the root system mass in combination with both Nerello Mascalese and with Nero d'Avola compared to 34 E.M., could represent a useful adaptation for obtaining soil water or nutrient resources when these are limited.

The lowest value of root system mass/above-ground mass was observed in both cultivars grafted to 34 E.M. and in both self-rooted cultivars. For the 34 E.M. rootstock, generally not considered resistant to drought conditions (Carbonneau 1985) and, for the self-rooted vines, this may indicate that under drier conditions the vines can obtain the limiting resources in a smaller soil mass ensuring a normal vegetative growth and good production, compared with the other root systems. Despite the roots from self-rooted vines being much fewer in number, the root system mass was similar to that of 34 E.M. due to a difference in root size. In particular the ≥2 mm root class was high for Nero d'Avola at the profile 0 from 41 to 100 cm depth. Moreover, although all the measured values of top and root growth for the self-rooted vines were lower than that for the grafted vines, the root system mass/above-ground mass ratio was similar indicating that an equilibrium was established between the below-ground and above-ground parts of the vines.

These vines produced a reasonable yield with good fruit TSS. The 34 E.M. root/shoot relation was similar to the root mass/shoot mass ratio for the Nerello Mascalese combination, while for Nero d'Avola the scion role was greater in combination with the 1103 P. rootstock.

Our results are in contrast with the findings of Williams and Smith (1991) who reported no effect of rootstock on the partitioning of dry matter among the organs, with the exception of the trunk. Our results indicate the yield was greatly influenced by the rootstock. In particular, the 34 E.M. rootstock, considered less vigorous than the others, showed a high yield when grafted to both scions due to the reduction in vegetative growth. When grafted to Nerello Mascalese, however, 34 E.M. musts exhibited lower TSS and higher TA than for 140 Ru. and 1103 P.; these differences may be a result of the 30% higher yield.

Conclusions

A comparison of self-rooted and three rootstocks of grapevines, each grafted with two scion cultivars, revealed significantly less growth for the self-rooted vines compared with that of the grafted vines. No evidence of phylloxera was observed in any self-rooted vines. Generally, root production in the heavy soil was maximum at 41–60 cm with fewer roots at the surface and lower depths. The results showed that the scion affected root growth, development and distribution. The scion affected most developmental parameters, such as the diameter of the root systems, the root density at 21–80 cm depth and the ratio of fine to coarse roots. These observations are broadly in line with those of other studies under similar environmental conditions.

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Table 4. Effect of root system on vine performance and yield components of Nerello Mascalese and Nero d'Avola†.

Cultivar	Root system	Yield (kg/vine)	Bunches/ vine (No.)	Bunch mass (kg)	Pruning mass (kg)	TSS (°Brix)	pH	TA (g/L)
Nerello Mascalese	34 Ecole de Montpellier	7.6 ± 1.1B	11.6 ± 3.1c	0.66 ± 0.2	2.05 ± 0.6 cd	20.4 ± 0.5C	3.2 ± 0.1	9.2 ± 0.6AB
	140 Ruggeri	13.5 ± 0.9A	14.9 ± 5.4ab	0.91 ± 1.3	2.22 ± 0.7bcd	22.2 ± 0.3A	3.1 ± 0.2	8.7 ± 0.5 BC
	1103 Paulsen	6.6 ± 0.7B	15.4 ± 5.4a	0.43 ± 0.2	2.93 ± 0.9abc	22.1 ± 0.1A	3.4 ± 0.1	8.6 ± 0.2 BC
	Self-rooted	6.1 ± 0.3 BC	15.6 ± 3.8a	0.39 ± 0.1	1.78 ± 0.5d	22.5 ± 0.3A	3.3 ± 0.2	9.5 ± 0.3A
Nero d'Avola	34 Ecole de Montpellier	6.0 ± 0.2 BC	12.0 ± 4.1bc	0.50 ± 0.2a	3.29 ± 0.8a	22.3 ± 0.4A	3.1 ± 0.2	8.0 ± 0.2DE
	140 Ruggeri	6.0 ± 0.6 BC	14.9 ± 3.2ab	0.40 ± 0.1	3.66 ± 0.6a	20.4 ± 0.5C	3.1 ± 0.2	8.2 ± 0.2CD
	1103 Paulsen	5.0 ± 0.7C	12.6 ± 4.9bc	0.39 ± 0.1	3.11 ± 0.9ab	21.2 ± 0.3B	3.3 ± 0.2	8.6 ± 0.2 BCD
	Self-rooted	4.7 ± 0.4C	16.3 ± 3.0a	0.29 ± 0.1	2.89 ± 0.4abc	22.6 ± 0.3A	3.2 ± 0.1	7.5 ± 0.3E
n		10	10	10	10	10	10	10
P-value		***	*	ns	*	***	ns	***

†Values are means of 10 years of analyses ± SD. Mean values in the same column indicated by different letters are significantly different at $P \leq 0.05$ level (*, lowercase letters) and at $P \leq 0.001$ (***, uppercase letters) based on Tukey's HSD test for factorial ANOVA.

Acknowledgement

We thank Professor Michela Centinari for a critical evaluation of the manuscript.

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Manuscript received: 22 January 2019
Revised manuscript received: 31 August 2019
Accepted: 25 September 2019

Supporting information

Additional supporting information may be found in the online version of this article at the publisher's website: http://onlinelibrary.wiley.com/doi/10.1111/ajgw.12415/abstract.

Figure S1. The root system of Nerello Mascalese on rootstocks (a) 34 Ecole de Montpellier, (b) 1103 Paulsen, (c) 140 Ruggeri and (d) self-rooted at the end of the study. The roots from nearby vines are painted white.

Figure S2. The root system of Nero d'Avola on rootstocks (a) 34 Ecole de Montpellier, (b) 1103 Paulsen, (c) 140 Ruggeri and (d) self-rooted at the end of the study. The roots from nearby vines are painted white.

Table S1. Main effects and interactions of cultivar, root system, profile and depth on root system performance.

Table S2. Main effects and interactions of cultivar and root system on the vegetative parameters of the vines.

Table S3. Main effects and interactions of cultivar and root system on yield components.

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