Root system distribution and yield of 'Conilon' coffee propagated by seeds or cuttings

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Abstract – The objective of this work was to evaluate the root system distribution and the yield of 'Conilon' coffee (*Coffea canephora*) propagated by seeds or cuttings. The experiment was carried out with 2x1 m spacing, in an Oxisol with sandy clay loam texture. A randomized complete block design was used, following a 2x9x6 factorial arrangement, with two propagation methods (seeds and cuttings), nine sampling spacings (0.15, 0.30, 0.45, 0.60, 0.75, and 0.90 m between rows, and 0.15, 0.30, and 0.45 between plants within rows), six soil depths (0.10–0.20, 0.20–0.30, 0.30–0.40, 0.40–0.50, and 0.50–0.60 m), and six replicates. Soil cores (27 cm^3) with roots were taken from 12 experimental units, 146 months after planting. The surface area of the root system and root diameter, length, and volume were assessed for 13 years and, then, correlated with grain yield. The highest fine root concentration occurred at the superficial soil layers. The variables used to characterize the root system did not differ between propagation methods. Moreover, no differences were observed for net photosynthetic CO_2 assimilation rate, stomatal conductance, internal CO_2 concentrations, and instantaneous water-use efficiency in the leaves. Cutting-propagated plants were more productive than seed-propagated ones.

Index terms: *Coffea canephora*, root diameter, root length, root volume, vegetative propagation.

Distribuição do sistema radicular e produtividade do café 'Conilon' propagado por sementes ou estaquia

Resumo – O objetivo deste trabalho foi avaliar a produtividade e a distribuição do sistema radicular do café 'Conilon' (*Coffea canephora*) propagado por sementes ou estaquia. O experimento foi realizado no espaçamento 2x1 m, em Latossolo com textura franco-argilo-arenosa. Utilizou-se o delineamento experimental de blocos ao acaso, em arranjo fatorial 2x9x6, com dois métodos de propagação (semente e estaca), nove espaçamentos (0,15, 0,30, 0,45, 0,60, 0,75 e 0,90 m, nas entrelinhas, e 0,15, 0,30 e 0,45 entre plantas), seis profundidades (0,10–0,20, 0,20–0,30, 0,30–0,40, 0,40–0,50 e 0,50–0,60 m) e seis repetições. Foram retirados monólitos (27 cm^3) de solo com raízes de 12 unidades experimentais, 146 meses após o plantio. A área superficial do sistema radicular bem como o diâmetro, o comprimento e o volume das raízes foram avaliados por 13 anos e correlacionados à produtividade de grãos. A maior concentração de raízes mais finas ocorreu nas camadas superficiais do solo. As variáveis utilizadas para caracterizar o sistema radicular não diferiram entre os métodos de propagação. Além disso, não se observaram diferenças quanto à taxa de assimilação fotossintética líquida de CO_2, nem quanto à condutância estomática, às concentrações internas de CO_2 e à eficiência do uso da água nas folhas. Plantas propagadas por estaquia foram mais produtivas do que as propagadas por sementes.

Termos para indexação: *Coffea canephora*, diâmetro de raízes, comprimento de raízes, volume de raízes, propagação vegetativa.

Introduction

Seed-derived plants from *Coffea canephora* Pierre ex A. Froehner are both genetically and morphologically heterogeneous, which results in substantial variations in plant architecture, disease resistance, maturation time, seed size and shape, and yield (Conagin & Mendes, 1961; Bragança et al., 2001). Therefore, commercial propagation of the species has been done by cuttings, which overcomes the variability caused by seed propagation. Cutting-propagation is physiologically viable and ensures maximum crop homogeneity, besides other desirable traits, especially grain maturation, fruit yield and size, and plant vigor

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(Weigel & Jurgens, 2002; Paiva et al., 2012). Moreover, this technique has allowed the establishment of coffee crops in areas with biotic or abiotic limitations (Miranda et al., 2011; Paiva et al., 2012).

Commercial 'Conilon' coffee fields use seedlings from cuttings. However, there are important questions related to the development of plants derived from cuttings, as for their ability to grow in depth and to anchor and support adult plants.

Root system characteristics are known to differ according to species, genotype, plant age, season, climate, plant density, root diameter, biotic stresses, and soil texture and structure (Lynch, 1995). Current studies addressing the characteristics of root system of coffee plants are still inconclusive as to its distribution in the soil profile, as well as to root physiology, size, and volume (Rena & Guimarães, 2000; Carvalho et al., 2008; Andrade Júnior, 2013). Therefore, additional knowledge on coffee root system can improve crop management, since a well-developed root system can enhance water and nutrient uptake and, also, increase the efficiency of soil-applied fungicides and insecticides, which can directly affect plant yield and tolerance to drought and other stresses (Franco & Inforzato, 1946; Rena & Guimarães, 2000; Carvalho et al., 2008).

The objective of this work was to evaluate the root system distribution and the yield of 'Conilon' coffee (*Coffea canephora*) propagated by seeds or cuttings.

Materials and Methods

This experiment was carried out in Vila Valério (18°57'S; 40°18"W, at approximately 150 m altitude), in the north-western portion of the state of Espírito Santo, Brazil. The climate type in the region is tropical, with hot and humid summers and dry winters. The annual rainfall is 1,200 mm, and precipitation volume concentrates between November and January. Annual average temperature is 23°C, with 29°C maximum mean and 18°C minimum mean (Comitê de Bacia Hidrográfica do Rio Doce, 2013). The soil in the area was classified as Typic Hapludox (Latossolo Vermelho Amarelo, Santos et al., 2013), with sandy clay loam texture, whose physical and chemical attributes (Silva, 2009) are described in Table 1.

The crop was established on November 22, 1999, in a 2x1 m spacing, following east-west orientation. The experiment was carried out in a randomized complete block design in a 2x9x6 factorial arrangement, with two forms of propagation (seed and cuttings), nine soil sampling spacing distances (15, 30, 45, 60, 75, and 90 cm between rows, and 15, 30, and 45 cm between plants in the rows) – which represented six samples in the career direction, and three between plants –, six soil depths (10–20, 20–30, 30–40, 40–50, and 50–60 cm), and six replicates. Each plot consisted of five plants with 12 replicates, until the 2006 harvest, and six replicates until the last harvest (2013). The number of replicates was reduced because some plants were cut off for visual assessment of the root system. In the place of cut plants, cacao plants were grown. In order to avoid interference of cacao plants on the results, only 50% of the plants that remained in the undisturbed areas were evaluated.

Conventional fertilizers were used from 2002 to 2006, when they were changed to organic fertilizers. After 2006, chemical fertilizers were applied only when soil analyses indicated possible nutrient deficiency. Crop management included irrigation.

Seed- and cutting-propagated plants were harvested 13 times during the growing seasons from 2001 (17-month-old plants) to 2013 (161-month-old plants). Total yield was estimated (kg ha^{-1}) according to individual yields. Harvested coffee at 5.33 L was considered to provide 1 kg of processed coffee, according to observations in the field.

Net CO_2 assimilation rate (A), stomatal conductance to water vapor (g_s), internal CO_2 concentration (Ci), and transpiration rates (E) were measured in two leaves, on 12 plants per treatment, using a portable Irga open system Ciras 2, (PP Systems, England). Measurements were taken between 8:00 and 10:00 h. During this interval, plants were subjected to field variations, ranging approximately between: 1,200 and 1,400 $\mu mol\ m^{-2}s^{-1}$ irradiance; 65 and 70% relative humidity; and 22 and 24ºC; with 380 $\mu L\ L^{-1}$ air CO_2 concentration. In general, maximum ranges occur by mid-morning (Silva et al., 2004). Determinations were performed in recently matured leaves, from the upper parts of plants.

Chlorophyll (Chl) fluorescence and gas exchange were evaluated on the same leaves, at the same times, using a Ciras 2, PP Systems. The photochemical quenching (qP) (Kooten & Snell, 1990) and the photosystem II (PSII) energy conversion efficiency

(Fv'/Fm') (Krüpa et al., 1993) were obtained under steady-state photosynthetic conditions (irradiance of 650 µmol m^{-2} s^{-1} and saturating flashes of 6,000 µmol m^{-2} s^{-1}).

In order to evaluate the root system, soil cores were taken from 12 experimental units, 146 months after planting, as follows: six plots with plants derived from seed, and six from cuttings (six replicates). The large number of treatments (2x9x6) and replicates (six plots per treatment) were able to provide a broad knowledge of the root system distribution of 'Conilon' coffee, and made it possible to build maps based on reliable results. Sampled monoliths had 27 cm^3 volume (3x3x3 cm). Samples were stored in plastic bags and refrigerated at approximately -10°C until analysis. The monoliths were washed to separate roots from the soil with running water and 30-mesh sieve. Roots retained in the sieve were transferred to another sieve (60 mesh) and were washed in water again.

Table 1. Soil chemical and textural properties, at six depths, after 146 months of 'Conilon' crop establishment.

Variable	Soil depth (cm)					
	0–10	10–20	20–30	30–40	40–50	50–60
pH	5.7	5.4	5.4	5.4	5.2	5.1
P (mg dm^{-3})	28	5	3	2	1	1
K (mg dm^{-3})	101	60	43	34	28	24
S (mg dm^{-3})	7	6	7	13	22	26
Ca ($cmol_c$ dm^{-3})	2.0	1.4	1.2	1.0	0.9	0.7
Mg ($cmol_c$ dm^{-3})	0.5	0.3	0.2	0.2	0.2	0.1
Al ($cmol_c$ dm^{-3})	0.0	0.3	0.3	0.3	0.4	0.4
H+Al ($cmol_c$ dm^{-3})	2.6	2.1	2.0	2.0	1.8	2.1
Na (mg dm^{-3})	49	34	27	26	25	22
SB ($cmol_c$ dm^{-3})	2.8	1.9	1.5	1.3	1.2	0.9
E ($cmol_c$ dm^{-3})	5.4	4.0	3.5	3.3	3.0	3.0
CEC ($cmol_c$ dm^{-3})	2.8	2.2	1.8	1.6	1.6	1.3
OM (dag kg^{-1})	1.9	1.1	0.9	0.8	0.7	0.5
m (%)	0	14	17	19	25	32
V (%)	51.5	46.9	43.0	39.2	39.4	29.1
Fe (mg dm^{-3})	74	108	125	121	125	110
Cu (mg dm^{-3})	0.5	0.3	0.3	0.2	0.2	0.2
Zn (mg dm^{-3})	6.7	2.4	0.6	0.2	0.2	0.2
Mn (mg dm^{-3})	42	35	33	20	9	7
B (mg dm^{-3})	0.82	0.77	0.47	0.30	0.27	0.23
Total sand (g kg^{-1})	610	586	582	520	524	500
Silt (g kg^{-1})	150	154	138	160	156	140
Clay (g kg^{-1})	240	260	280	320	320	360
Soil density (g cm^{-3})	1.06	1.25	1.51	1.51	1.65	1.65

SB, sum of bases; E, cation exchange capacity at pH 7.0; CEC, effective cation exchange capacity; OM, organic matter; m, aluminium saturation index; V, base saturation index.

The washed roots were photographed with a digital camera and, subsequently, they were analyzed using the Safira software, which is an analysis system for fibres and roots (Embrapa Instrumentação Agropecuária, São Carlos, SP, Brazil) (Jorge & Rodrigues, 2008), in order to quantify length, volume, surface area, and diameter of roots. Fine root content per volume (cm^3) of soil was estimated with roots with less than 1 mm diameter.

Nonnormally distributed data associated with root diameter, surface area, length, and volume were transformed [y = Log (x + 1)] prior to the analysis of variance and comparison of means; notwithstanding, the original data were shown.

Statistical analysis was performed using Assistat 7.6 (Silva, 2012). Data were subjected to the analysis of variance, following a Box-Cox transformation. Means were compared by Tukey's test, at 5% probability.

Isoline diagrams were drawn using the GS+ version 7 software (Gamma Design Software, Plainwell, Michigan, USA), in order to illustrate the spatial distribution of the root system in the soil profile, in two dimensions.

Results and Discussion

The yield of cutting-propagated plants was significantly higher in the 1st, 2nd, 4th, 8th, 10th and 13th harvests (Table 2), whereas seed-propagated plants did not show significantly higher yields in none of the studied years. The highest yield difference between the two groups of plants was found after the first two harvests. The overall production along the experimental period was 43,788 kg ha^{-1} for cutting- propagated plants, and 35,644 kg ha^{-1} for seed-propagated ones. Additionally, the average annual yields for the 13-year period were respectively 3,368 and 2,742 kg ha^{-1}. Therefore, cutting-propagated plants had a significantly higher yield during the trial, showing a clear advantage over the seed-propagated ones.

Plants derived from cuttings had earlier production capability, which might be caused by the greater physiological maturity of these individuals. In that respect, these plants were able to develop a higher number of productive (plagiotropic) branches sooner than seed-derived ones. This might be related to the fact that 'Conilon' clones are produced from cuttings of physiologically differentiated tissue (Bragança et al., 2001). In contrast, seed-derived plants are able to develop plagiotropic (i.e., productive) branches only by the ninth or tenth leaf axil. This difference delays fruiting stage and the full production potential of plants, which was clearly observed in the first four years of this experiment. These findings agree with Carvalho et al. (2011), who observed that *C. arabica* 'Catuaí' plants had a better initial development when derived from vegetative propagation (somatic embryogenesis). However, the higher yields observed for plants derived from cuttings may also have been caused by the better genetic material used for the propagation of these plants.

Net photosynthetic CO_2 assimilation, stomatal conductance to water vapor, internal CO_2 concentrations, and instantaneous water-use efficiency did no differ between propagation methods (Table 3). Moreover, the energy harvesting efficiency and photochemical quenching were also statistically similar.

Regarding the root system, the average diameter of lateral roots was significantly higher in plants derived from seed (Table 4 and Figure 1), mostly due to data variation in depth, since no differences were observed in the horizontal position (distance from the coffee tree trunk). Seed-grown plants had a significantly greater diameter in the 50–60 cm layer, which was similar to that reported for *C. arabica* plants (Rena & Guimarães, 2000).

When comparing root concentrations according to the horizontal distance from the trees, the first centimetres of the top-soil layer had greater values of root length and volume between rows than the ones observed within rows (Figure 2). The area between plants in the rows did not receive topdressing fertilization. Therefore, this result most likely occurred because of the higher fertility in the space between rows (Sousa et al., 2002).

Root surface area, length, and volume did not differ between the propagation methods in none of the evaluated distances or depths (Table 5 and Figure 2). Therefore, under the particular conditions of this study, the root system of 'Conilon' coffee was not affected by the propagation method. This finding contrasts with the results reported for *C. arabica*, in which total root length and dry weight of cutting-propagated plants were greater than those of seed-propagated ones (Jesus et al., 2006).

No interaction was observed between the evaluated distances and depths, for data related to root surface area, length, and volume. Roots had a higher concentration in the top soil layer (0–10 cm), which decreased with depth (Table 6). Roots had lower concentrations in the layers below 40 cm, showing a parallel with nutrient concentrations (P, K, and Ca) (Table 1). The spatial distribution of roots in the soil profile confirmed these trends (Figures 2). Mota et al. (2006) reported the highest concentration of coffee roots up to 0.20 m depth, and Rodrigues et al. (2001) observed lower root concentrations at deeper soil layers. This behavior of

Table 2. Yield of *Coffea canephora* 'Conilon' propagated by seed or cuttings, from 2001 (17 months) to 2013 (161 months)[(1)].

Propagation	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010	2011	2012	2013
Cutting	421a	5,795a	1,629a	3,135a	1,562a	4,325a	2,330a	4,349a	2,861a	4,410a	3,812a	4,051a	5,107a
Seed	73b	3,220b	1,269a	2,355b	1,549a	3,745a	2,513a	3,446b	2,977a	3,099b	4,051a	4,170a	3,178b
CV (%)	50.2	16.9	35.3	26.2	34.9	27.4	22.2	26.1	45.6	17.66	33.23	26.26	30.04

[(1)]Means followed by equal letters, in the columns, do not differ, by Tukey's test, at 5% probability.

Table 3. Net photosynthesis (A), stomatal conductance to water vapor (gs), internal CO_2 concentration (Ci), transpiration (E), and two parameters for chlorophyll fluorescence (Fv'/Fm', qP) of *Coffea canephora* 'Conilon' plants propagated by seed or cuttings.

Propagation	A (μmol m^{-2} s^{-1} CO_2)	Gs (mmol m^{-2} s^{-1} H_2O)	Ci (μmol mol^{-1})	E (mmol m^{-2} s^{-1} H_2O)	Fv'/Fm'	qP'
Cutting	13.88a	85.12a	230.58a	1.76a	0.60a	0.49a
Seed	13.01a	98.02a	227.17a	1.59a	0.62a	0.49a
CV (%)	13.88	39.04	27.51	15.02	6.45	28.99

[(1)]Means followed by equal letters, in the columns, do not differ, by Tukey's test, at 5% probability. Fv'/Fm', photochemical efficiency of PS II under photosynthetic steady-state conditions; qP, photochemical quenching.

root systems can also be observed for other species, such as passion fruit (Sousa et al., 2002; Lucas et al., 2012) and orange rootstocks (Zaccheo et al., 2012).

Although root development in coffee is mainly related to the genetic characteristics of the plant (Martins et al., 2013), other factors may also interfere in the spatial distribution, such as the propagation form, seedling stage, and amount of water in the soil (Franco & Inforzato, 1946). The availability of soil nutrients to plants can also influence the deep root density and distribution (Bakker et al., 2006). According to Witschoreck et al. (2003), the concentration of fine roots in the soil surface layer is related to the higher concentrations of organic matter and nutrients, and to favorable physical conditions. Nutrient availability can be a limiting factor for coffee root development, particularly for Brazilian soils (Rena & Guimarães, 2000).

Table 4. Root diameter (mm) of *Coffea canephora* 'Conilon' propagated by seed or cuttings at different soil depths[(1)].

Soil depth (m)	Cuttings	Seed
0–10	0.659Aa	0.687Aa
10–20	0.704Aa	0.700Aa
20–30	0.770Aa	0.741Aa
30–40	0.679Aa	0.703Aa
40–50	0.636Aa	0.673Aa
50–60	0.415Bb	0.592Ba
CV (%)	37.89	32.45

[(1)]Means followed by equal letters, lowercase in the rows and uppercase in the columns, do not differ by Tukey's test, at 5% probability.

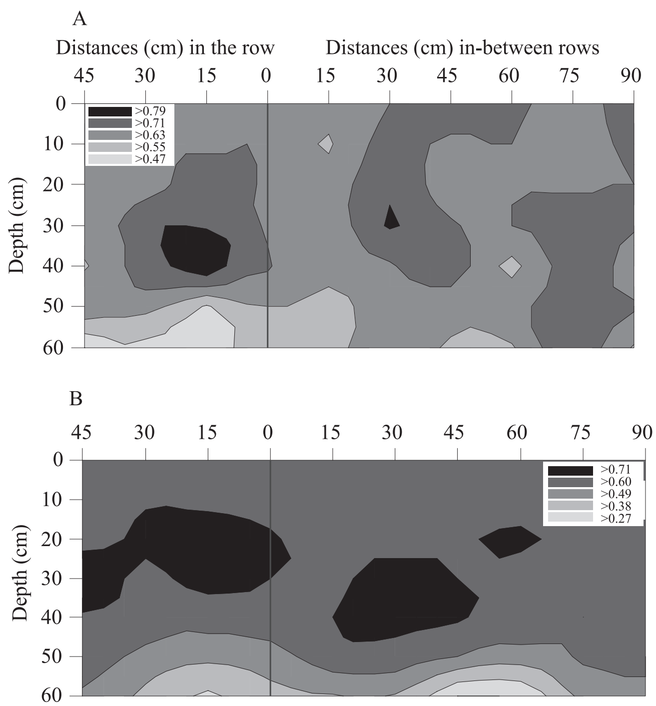

Figure 1. Spatial distribution of the root diameters (mm) of *Coffea canephora* 'Conilon', propagated by seed (A) or cuttings (B), at different horizontal distances and depths. The zero point refers to the plant location.

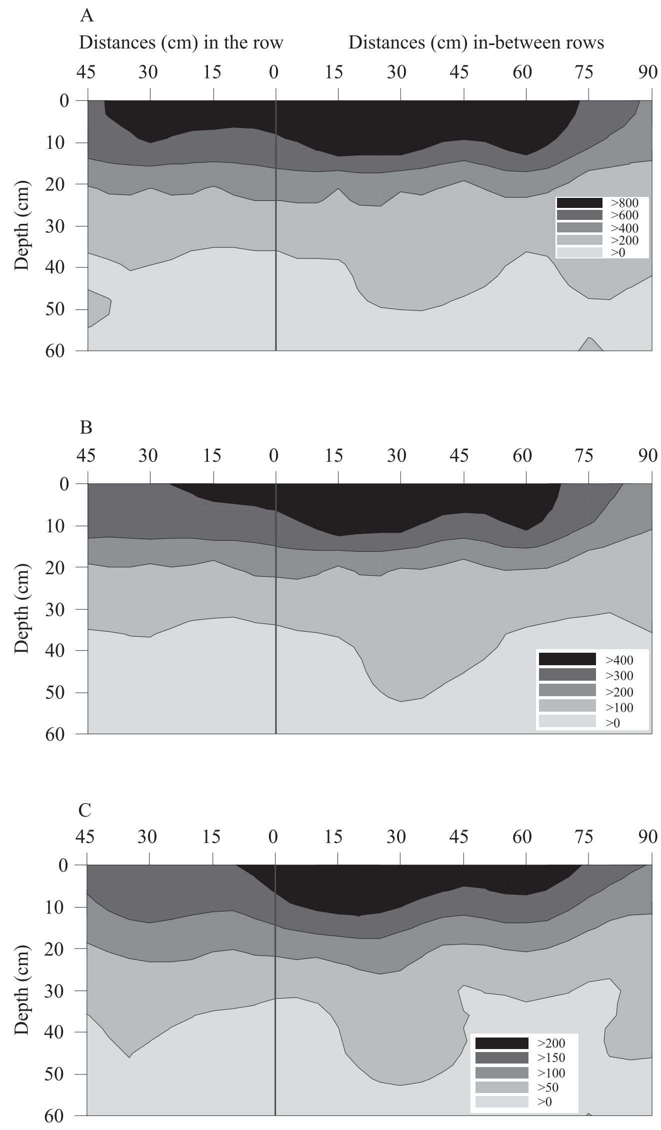

Figure 2. Spatial distribution of: A, root surface area (cm^2 dm^{-3}); B, root length (m dm^{-3}); and C, root volume (mm^3) for *Coffea canephora* 'Conilon' propagated by seed or cuttings, at different horizontal distances and depths. The zero point refers to the plant location.

Table 5. Surface area, length, and volume of *Coffea canephora* 'Conilon' roots propagated by seed or cuttings, at different spacing distances and depths.

Soil depth (cm)	Distances between rows (cm)						Distances within rows (cm)		
	15	30	45	60	75	90	15	30	45
	Surface area (cm² dm⁻³)								
0–10	977.2	974.4	773.1	952.1	651.6	509.9	757.1	828.4	724.9
10–20	404.7	414.2	343.1	472.4	271.6	247.7	394.0	409.6	393.0
20–30	377.5	384.8	187.6	284.2	226.3	291.4	297.9	323.8	303.2
30–40	156.2	402.0	307.7	148.6	265.5	206.1	113.4	193.0	149.5
40–50	167.9	194.2	151.4	143.7	184.9	172.4	95.1	182.2	251.3
50–60	104.7	112.7	97.1	111.3	219.4	133.8	88.9	146.3	139.6
	Length (m dm⁻³)								
0–10	471.5	445.7	357.4	435.7	303.1	210.8	368.5	356.8	346.2
10–20	177.1	191.7	156.9	199.4	125.2	114.4	165.5	179.7	180.6
20–30	171.8	152.8	79.5	129.0	106.7	122.8	116.4	139.5	127.0
30–40	68.2	174.9	134.0	66.1	112.8	85.7	44.7	85.3	73.7
40–50	66.6	115.7	67.0	57.2	82.8	88.2	37.6	57.1	103.9
50–60	31.2	61.3	49.7	46.9	91.9	61.5	32.6	59.3	63.8
	Volume (mm³)								
0–10	244.6	207.1	160.7	197.1	137.9	119.3	148.6	187.1	141.2
10–20	87.6	113.7	74.0	110.2	57.9	52.3	97.2	100.8	84.4
20–30	86.6	98.6	42.5	59.2	45.0	74.5	76.7	77.0	74.0
30–40	35.3	100.2	68.5	30.7	61.6	51.1	28.9	45.3	27.4
40–50	42.3	58.7	33.9	42.0	40.2	43.6	24.7	60.6	60.2
50–60	30.1	25.7	18.1	28.5	51.3	26.7	25.6	34.2	28.5

Table 6. Surface area, length, and volume of roots of *Coffea canephora* 'Conilon' propagated by seed or cuttings at different soil depths.

Depth (cm)	Surface area (cm² dm⁻³)	Length (m dm⁻³)	Volume (mm³)
	Absolute values		
0–10	794.3a	366.2a	171.5a
10–20	372.3b	165.6b	86.4b
20–30	297.4bc	127.3bc	70.5bc
30–40	215.8cd	93.9cd	49.9cd
40–50	174.4cd	75.1cd	45.1cd
50–60	128.2d	55.4d	29.9d
CV (%)	37.71	37.89	39.28
	Percentual values		
0–10	40.13	41.45	37.83
10–20	18.81	18.75	19.06
20–30	15.03	14.41	15.55
30–40	10.90	10.63	11.00
40–50	8.66	8.50	9.94
50–60	6.84	6.27	6.59

(1)Means followed by equal letters, in the columns, do not differ, by Tukey's test, at 5% probability.

Conclusions

1. Cutting-propagated plants of *Coffea canephora* 'Conilon' are more productive than seed-propagated ones.

2. Surface area, length, and volume of 'Conilon' roots are not affected by the propagation method, and show higher values in the soil surface, in-between the rows.

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