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Rootstocks for high fruit yield and quality of 'Tahiti' lime under rain-fed conditions

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ABSTRACT

Despite considerable research conducted on 'Tahiti' lime [*Citrus latifolia* (Yu Tanaka) Tanaka] in several countries, few long-term studies have focused on rootstock effects on fruit production and quality under non-irrigated conditions. As for many other fruit crops, rootstock studies for 'Tahiti' lime are often based on the evaluation of several horticultural responses simultaneously, instead of considering multivariate statistical approaches which may provide with more comprehensive information. Consequently, a trial was installed to evaluate the horticultural performance of non-irrigated 'Tahiti' lime trees budded onto the following 12 rootstocks: 'HRS 801' and 'HRS 827' hybrids; 'Rubidoux', 'FCAV' and 'Flying Dragon' trifoliates; 'Sun Chu Sha Kat' and 'Sunki' mandarins; 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' limes; 'Carrizo' citrange, 'Swingle' citrumelo, and 'Orlando' tangelo. The trial was installed in 2001, in an 8 m × 5 m spacing with no supplementary irrigation. Measurements of yield, fruit quality oriented to different consuming markets, canopy volume and tree tolerance to drought, were performed. A multivariate cluster analysis identified both 'Rangpur' lime rootstocks as those inducing larger cumulative yield and higher percentage of fruits for the domestic market, with highest drought tolerance to the trees. Despite of their high susceptibility to drought stress under non-irrigated conditions, the 'Flying Dragon' and 'FCAV' trifoliate rootstocks performed outstandingly for 'Tahiti' lime, inducing higher yield efficiency, early bearing and larger percentage of high-quality fruits for foreign markets, with smaller trees more suitable for high-density plantings.

1. Introduction

'Tahiti' lime, also known as Persian lime or Bearss lime, is a triploid hybrid largely grown in India, Mexico, China, Argentina, and also in Brazil, where it occupies a cultivated area of 43,000 ha producing more than 965,000 t per year (IBRAF, 2011). The State of São Paulo is the main producer, with 28,000 ha (64% of the total planted area) and over 750,000 t (77% of the national production) produced mainly under rain-fed subtropical conditions. Nearly 83% of the Brazilian 'Tahiti' lime production is consumed by the domestic market as fresh fruit, while 12% of the annual harvest is used to produce concentrated juice, essential oil, and pectin that supply the pharmaceutical, food and perfumery industries (ABPEL, 2011). Despite significant increases in recent years, Brazilian exports of fresh 'Tahiti' limes represent just 5% of the overall production, being mainly shipped to Holland, United Kingdom, Germany and Canada (ABPEL, 2011).

In Brazil, 'Tahiti' limes continuously bloom throughout the year, with a total of three to five harvests per year in the State of São Paulo (Espinoza-Nuñez et al., 2011). The main harvest period occurs in the first semester of the year, representing over 70% of the annual harvest, while a secondary harvest, of nearly 30% of the annual fruit production, is registered in the second semester (Figueiredo et al., 2003). Thus, different marketing strategies are applied for fresh fruit produced along the year, either to promote the exports, mainly destined to European markets, during the first semester, or to supply the domestic market in the second semester during the off season, when highest prices are registered. Consequently, different quality standards are applied to fresh fruits harvested along the year, based on the particular target market to be reached.

In Brazil, 'Tahiti' limes are mainly grown under non-irrigated conditions by small family farmers, with low yields, of around 20 t ha^{-1}, resulting from large plant spacing and short grove lifespan that reduce crop profitability (Silva et al., 2008).

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Several studies have highlighted the necessity of rootstock diversification for increasing fruit yield and quality of 'Tahiti' lime under rain-fed conditions (Figueiredo et al., 2003; Stenzel and Neves, 2004; Stuchi et al., 2003). Nonetheless, rootstock selection is mainly based on few horticultural variables, instead of considering multiple agronomical responses commonly measured in this kind of studies. The objective of this work was to evaluate the response of 'Tahiti' trees budded onto 12 rootstocks under rain-fed subtropical conditions in São Paulo State, Brazil, with emphasis on rootstock influence over plant size, plant tolerance to drought and fruit yield and quality for the domestic and foreign markets. A multivariate clustering analysis technique was utilized to identify aggregates of rootstocks that induced similar characteristics to 'Tahiti' lime trees, considering 12 evaluated variables that were significantly affected by the rootstocks.

2. Materials and methods

2.1. Field trial and plant material

The experimental grove was planted in February 2001 at the Estação Experimental de Citricultura de Bebedouro, São Paulo State, Brazil (20°53′16″S latitude, 48°28′11″W longitude, 680 m altitude), from 12 month-old plants (nursery trees) disposed in a 8 m × 5 m spacing (250 plants ha^{-1}), on a total area of 2880 m^2. The climate is Köeppen's Cwa, with a mean temperature of 23.5 °C and an annual rainfall of 1522 mm. The soil is classified as Haplustox (38% clay) (EMBRAPA, 1999). Plants were formed from buds of the 'IAC 5-1' 'Tahiti' lime selection collected at the Active Citrus Germplasm Bank of the Centro APTA Citros Sylvio Moreira in Cordeirópolis, São Paulo State that were grafted onto each one of the following 12 rootstocks (treatments): the hybrids 'HRS 801' ['Changsha' mandarin (*Citrus reticulata* Blanco) × *Poncirus trifoliata* 'English Small'] and 'HRS 827' ['Rangpur' lime (*Citrus limonia* Osbeck) × 'Swingle' trifoliate (*P. trifoliata* 'Swingle')]; the trifoliates (*P. trifoliata* (L.) Raf.) 'Rubidoux', 'FCAV', and 'Flying Dragon' (*P. trifoliata* var. *monstrosa*); the 'Sun Chu Sha Kat' mandarin (*C. reticulata* Blanco); the 'Sunki' mandarin (*Citrus sunki* (Hayata) hort. ex Tanaka); the 'Rangpur' limes (*C. limonia* Osbeck) 'Cravo Limeira' and 'Cravo FCAV'; 'Carrizo' citrange (*Citrus sinensis* (L.) Osbeck × *P. trifoliata*), 'Swingle' citrumelo (*Citrus paradisi* Macf. × *P. trifoliata*), and 'Orlando' tangelo (*C. paradisi* × *Citrus tangerina* cv. 'Dancy'). Rootstock seeds were obtained from plants grown at the Estação Experimental de Citricultura de Bebedouro, except for the 'HRS 801' and 'HRS 827' hybrids, 'Rubidoux' trifoliate and the 'Rangpur' lime 'Cravo Limeira' rootstocks, provided by the Centro APTA Citros Sylvio Moreira, and the 'Cravo FCAV' and 'FCAV' trifoliate rootstocks, obtained at the Universidade Estadual Paulista 'Júlio de Mesquita Filho' in Jaboticabal, São Paulo State. No supplemental irrigation was applied. Trees were not pruned and standard cultural practices for citrus were applied (Mattos Júnior et al., 2005).

2.2. Plant yield and size

Fruit yield of non-irrigated 'Tahiti' lime trees was recorded on each commercial harvest from the second through the seventh year after planting (2003–2008). Within each year, a mean of four harvests were recorded. For each treatment (rootstock), cumulative annual yield (2003–2008) was determined. The percentage of total cumulative yield recorded at the second commercial harvest (2004) was utilized to assess the rootstock effect on early bearing. Rootstock effect over the annual harvest distribution was inferred from the percentage of cumulative yield (2003–2008) separately picked in the first and second semesters.

Mean alternate bearing index (*ABI*) was calculated from the second through the seventh year after planting (2003–2008), by means of the following expression: $ABI = 1/(n-1) \times \{|a_2 - a_1|/(a_2 + a_1) + |a_3 - a_2|/(a_3 + a_2) + \ldots + |a_n - a_{n-1}|/(a_n + a_{n-1})\}$, where n = number of years and $a_1, a_2, \ldots, a_{n-1}, a_n$ = yield of the corresponding years (Stenzel and Neves, 2004).

Plant size was estimated by the canopy volume (*V*), as calculated from individual measurements of tree height (*H*) and width in parallel (D_l) and perpendicular (D_r) directions to the tree row recorded on May (2006 through 2008), assuming that the tree shape was one half prolate spheroid, by using the formula: $V = (\pi/6) \times H \times D_l \times D_r$ (Zekri, 2000). Yield efficiency (*YE*) was computed from the relationship between fruit yield (kg $plot^{-1}$) and canopy volume (m^3 $plot^{-1}$) (2006 through 2008). The relationship between mean *YE* and *V* values was analyzed by a regression model.

2.3. Plant tolerance to drought

Drought tolerance was evaluated during the winter dry period (August), by using two approaches: (i) visual assessment of canopy water stress of each tree from 2006 through 2008, based on a three-score rating scale (scores 1, 2 and 3 for low, medium and high tolerance to water stress, respectively), as utilized on previous field studies with citrus species conducted in the experimental site (Stuchi et al., 2000); and (ii) leaf color, measured in 2008 with a Minolta CR-300 colorimeter and estimated through the variables L^* (luminosity), C^* (chromaticity) and h° (hue or color angle), on samples of 20 leaves per treatment. L^* ranges from black (0) to white (100). C^* values closer to zero represent gray colors, while values closer to 60 express vivid colors. Hue (h°) angles closer to 180° represent greener colors, while values closer to 90° indicate yellowish color. Green leaf color was described by low L^*, low C^* and high h° values (McGuire, 1992). Leaf color measurement for estimating drought tolerance was formerly utilized in other citrus species by Deidda et al. (1996), being highly correlated with visual assessment of plant drought tolerance of 'Folha Murcha' sweet oranges as previously evaluated in the experimental site (Cantuarias-Avilés et al., 2011).

2.4. Fruit quality measurements

Quality determinations were performed separately on fruits destined to domestic and foreign markets. For the domestic market, fruits were collected in the main annual harvest during the first semester (February–March). Fruit quality was assessed from 2006 to 2008 (year 5–7 after planting) on samples of five fruits collected from each plot, and the following variables were measured: (i) fruit mass and size, assessed by the sample fresh weight and fruit equatorial diameter; (ii) juice content (*JC*, %), as calculated from the relationship between the weight of the extracted juice and the fruit sample weight; (iii) total soluble solids content (*TSS*), expressed in °Brix, by direct reading in a temperature-compensated digital refractometer, and (iv) titratable acidity (*TA*), expressed as percentage of citric acid, by titration of 25 ml of juice with 0.31 N NaOH.

At the main harvest on March 12th, 2007 the fruit picked from each plot was classified in a commercial packing-house line separately for domestic or foreign market, based on its external appearance (size, peel color and roughness and absence of injures and spots). On fruit classified for export, individual measurements of diameter and peel color were performed on samples of ninety fruits randomly collected from each treatment. Fruit diameter data were used to draw the carton size distribution according to the packing grades defined for 'Tahiti' limes exported to European markets (64, 60, 54, 48 and 42 fruits per 4.5 kg carton). Peel color was assessed at two opposing sides around the equatorial region of each

Table 1
Cumulative yield after 7 years planting (*CY*), percentage of cumulative yield in the second commercial harvest (*CY 2nd crop*), mean cumulative yield percentage harvested in the first (*PCYS1*) and second semester (*PCYS2*), alternate bearing index (*ABI*), canopy volume (*V*) and yield efficiency (*YE*) of non-irrigated 'Tahiti' lime trees budded onto 12 rootstocks.

Rootstock	*CY*[#,**] (t ha^{-1})	*CY 2nd crop*[##] (%)	*PCYS1*[§] (%)	*PCYS2*[§§] (%)	*ABI*[#,**]	*V*[*] (m^3)	*YE*[*,**] (kg m^{-3})
'Cravo FCAV' 'Rangpur' lime	124.8 a	18.0 abc	61.7	38.3	0.27 b	43.8 ab	1.84 ab
'Cravo Limeira' 'Rangpur' lime	111.8 a	17.6 abc	62.1	37.9	0.33 ab	37.4 abc	2.01 ab
'HRS 801' hybrid	104.7 ab	14.6 abc	55.4	44.6	0.41 ab	42.6 ab	1.94 abc
'HRS 827' hybrid	103.3 ab	13.7 abc	63.0	37.0	0.33 ab	36.3 abc	1.66 abc
'FCAV' trifoliate	85.7 abc	13.8 abc	58.1	41.9	0.37 ab	31.3 bc	1.98 ab
'Sunki' mandarin	82.0 abc	21.1 ab	64.1	35.9	0.27 b	37.4 abc	1.41 bc
'Carrizo' citrange	81.6 abc	12.4 bc	64.5	35.5	0.32 ab	43.5 ab	1.55 bc
'Rubidoux' trifoliate	67.3 bc	19.7 ab	50.7	49.3	0.38 ab	23.2 cd	2.40 ab
'Swingle' citrumelo	62.0 bcd	19.5 ab	61.3	38.7	0.34 ab	37.1 ab	1.25 bc
'Orlando' tangelo	56.7 bcd	13.2 bc	66.0	34.0	0.45 a	48.6 a	0.79 c
'Sun Chu Sha Kat' mandarin	51.1 cd	8.2 c	51.7	48.3	0.45 a	36.9 abc	1.27 bc
'Flying Dragon' trifoliate	38.8 d	22.6 a	58.1	41.9	0.40 ab	9.5 d	3.18 a
CV (%)	6.9	26.7			10.5	21.2	6.7
P-value	<0.0001	<0.0001			0.0012	<0.0001	<0.0001

Means followed by different letters in columns are significantly different at the indicated *P*-value (Fisher's test) by Tukey's test ($P < 0.05$).
[#] 2003–2008.
[##] 2004.
[§] January–June.
[§§] July–December.
[*] Mean values from 2006 to 2008.
[**] Original data were transformed ($\hat{y} = \log_{10} y$) to follow a normal distribution.

fruit by measuring the L^* (luminosity), C^* (chromaticity) and h° (hue or color angle) values with a Minolta CR-300 colorimeter (McGuire, 1992).

2.5. Experimental design and data analysis

The experiment was conducted following a randomized complete block design, with six replications and one plant per plot, with a total of 72 trees under evaluation. Parametric and nonparametric analyses were performed using the Fisher's and Friedman's tests, respectively. The nonparametric approach was used for ordinal variables (drought tolerance scores) and for variables that did not fit the normal distribution. Original data of cumulative yield, yield efficiency, alternate bearing index [$\hat{y} = \log_{10}(y)$], and the colorimetric variables L^* [$(\hat{y} = y^{-2})$] and C^* [$(\hat{y} = y^{-1})$] were transformed using the Box Cox method to follow a normal distribution. Comparisons among means were performed by the Tukey's and the Dunnett's test ($P < 0.05$ significance level). Data of 12 variables that were influenced by the rootstocks (cumulative yield, percentage of total cumulative yield assessed in the second commercial harvest, percentage of exportable fruits, alternate bearing index, cumulative yield percentage harvested in the second semester, yield efficiency, canopy volume variation, drought tolerance mean score, and fruit weight, diameter, *TSS* and *TA* in the fruit destined to the domestic market) were subjected to a multivariate cluster analysis, in order to classify rootstocks in groups of closer similarity, according to their general horticultural performance. Hierarchical cluster analysis was performed on standardized means of 12 variables by using the single linkage method, and a dendrogram was drawn (Manly, 2005).

3. Results

3.1. Plant yield and size

Cumulative annual yield of non-irrigated 7 year-old 'Tahiti' lime trees was significantly larger when budded onto 'Cravo FCAV' and 'Cravo Limeira' 'Rangpur' limes, followed by those grafted onto the 'HRS 801' and 'HRS 827' hybrids. On the other hand, lowest cumulative yields were recorded on trees budded onto the 'Flying Dragon' trifoliate (Table 1). Nonetheless, under rain-fed conditions, 'Tahiti' lime trees had largest cumulative yield percentage at the second commercial crop when budded onto the 'Flying Dragon' trifoliate, followed by 'Sunki' mandarin, 'Rubidoux' trifoliate and 'Swingle' citrumelo. Conversely, the lowest cumulative yields of 'Tahiti' lime trees at their second commercial crop were recorded onto the 'Sun Chu Sha Kat' rootstock.

For most of the rootstocks, larger percentages of cumulative yield after seven years from planting were recorded during the main harvest (February through March) in the first semester. However, 'Tahiti' lime plants grafted onto the 'Rubidoux' trifoliate and the 'Sun Chu Sha Kat' rootstocks had a more even yield distribution between both semesters (Table 1).

Until the seventh year after planting, higher *ABI* values were observed on 'Tahiti' lime trees budded onto the 'Orlando' tangelo and the 'Sun Chu Sha Kat' mandarin, while plants budded onto the 'Cravo FCAV' 'Rangpur' lime and the 'Sunki' mandarin showed the lowest *ABI* scores (Table 1).

Larger canopy volume for the 2006–2008 period (years 5–7 after planting) were observed on 'Tahiti' lime trees grafted onto the 'Orlando' tangelo, followed by those grafted onto 'Cravo FCAV' 'Rangpur' lime, 'Carrizo' citrange and 'HRS 801' rootstocks (Table 1). The smallest size of the plants grafted onto the 'Flying Dragon' trifoliate contrasted with their largest yield efficiency. From 2006 through 2008, mean values of yield efficiency and canopy volume of 'Tahiti' lime trees budded onto the different rootstocks showed a significant linear relationship ($P < 0.0017$) and were highly correlated ($r = 0.8247$) (Fig. 1). Under rain-fed conditions, the 'Flying Dragon' trifoliate performed remarkably as rootstock for 'Tahiti' lime in terms of productivity, by conferring the largest yield efficiency in the smallest canopy. On the other hand, large-sized 'Tahiti' plants grafted onto the 'Orlando' tangelo produced lowest fruit mass per unit canopy volume (Fig. 1).

3.2. Plant tolerance to drought

Higher mean drought tolerance scores between the fifth and the seventh year after planting (2006–2008) were recorded on 'Tahiti' plants grafted onto the 'Cravo FCAV' and 'Cravo Limeira' 'Rangpur' limes, followed by those grafted onto the 'HRS 801' hybrid

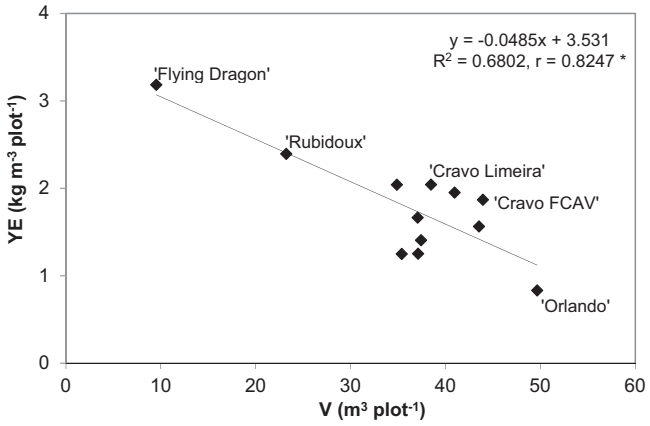

Fig. 1. Linear relationship between mean yield efficiency (*YE*) and mean canopy volume (*V*) of non-irrigated 'Tahiti' lime plots budded onto 12 rootstocks. Bebedouro, SP, Brazil, 2006–2008.

and the 'Sunki' mandarin. The 'Flying Dragon' trifoliate rootstock induced the lowest mean drought tolerance to non-irrigated 'Tahiti' lime trees during the winter dry season. The other rootstocks conferred moderate drought tolerance to the plants (Table 2). In 2008, all the colorimetric variables recorded in the driest month of the drought period (August) indicated a better leaf water status of 'Tahiti' plants grafted onto 'Cravo FCAV' and 'Cravo Limeira' 'Rangpur' limes and onto 'Sunki' mandarin and 'HRS 801' hybrid, whose leaves remained greener during the drought season. By the time of leaf colorimetric evaluation, a total of 151 mm of water deficit were recorded in the experimental site. Canopy water deficit was extreme on plants grafted onto the 'FCAV' and 'Flying Dragon' trifoliates, which showed a lighter and more yellowish leaf color, as expressed by higher L^* and C^* and lower h° values, confirming the lowest drought tolerance conferred by these rootstocks to the 'Tahiti' scion (Table 2).

3.3. Fruit quality

During the first semester 'Tahiti' lime trees budded onto the 'Flying Dragon' trifoliate yielded higher percentages of fruits classified as suitable for export (Table 3), and lower percentages of fruits for the domestic market. Trees budded onto the 'FCAV' trifoliate, 'Rubidoux' trifoliate and 'Sun Chu Sha Kat' rootstocks also yielded high percentages of export fruit. On the other hand, 'Tahiti' lime trees grafted onto the 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' limes yielded the lowest percentages of fruits for the foreign market, and larger percentages of fruits for the domestic market.

Among the fruit classified for export, higher percentages of large packing carton sizes were observed on 'Tahiti' trees grafted onto the 'Flying Dragon' trifoliate, while the lowest percentages of large-sized export fruits were recorded on trees budded onto 'Cravo FCAV' 'Rangpur' lime, 'Orlando' tangelo and the 'HRS 827' rootstocks (Table 3).

To stress the effects of different rootstocks on export fruit quality, the mean values of colorimetric variables describing peel color were compared by the Dunnet's test against those measured on fruits picked from trees grafted onto the 'Cravo Limeira' 'Rangpur' lime, the main rootstock in use for rain-fed 'Tahiti' lime production in Brazil (Table 3). Trees budded onto the 'Carrizo' citrange and the 'HRS 827' rootstocks produced fruits with lighter peel color (higher L^* values) than those harvested from trees on 'Cravo Limeira' 'Rangpur' lime, while the fruits picked from trees on 'Sunki' mandarin and 'Cravo FCAV' 'Rangpur' had darker peel color (lower L^* values). 'Tahiti' limes harvested from trees grafted on 'Sunki' mandarin, 'Swingle' citrumelo, 'HRS 801' hybrid and 'Sun Chu Sha Kat' mandarin had less vivid color (lower C^*), as compared to fruits picked from trees onto the 'Cravo Limeira' 'Rangpur' lime. Hue (h°) peel values of fruits from trees budded onto the 'Flying Dragon' trifoliate, 'FCAV' trifoliate and 'Cravo FCAV' 'Rangpur' lime were similar to those measured on fruits picked from trees onto the 'Cravo Limeira' 'Rangpur' lime, while all the other rootstocks induced higher hue values, associated to greener peel color (Table 3).

Mean values of variables describing internal fruit quality of 'Tahiti' limes picked during the first semester from the fifth through the seventh year after planting (2006–2008), indicated that the 'Rubidoux' and the 'Flying Dragon' trifoliate rootstocks induced larger fruit weight and highest juice *TSS* and *TA* values to this scion (Table 4). On the other hand, 'Tahiti' lime trees grafted onto the 'Orlando' tangelo yielded the smallest fruits, with lower *TSS* contents.

3.4. Rootstock effect on the overall tree performance

Considering a distance between rootstock clusters of 0.6, as calculated by the single-linkage method (Manly, 2005), the dendrogram of the multivariate cluster analysis depicted two different aggregates of rootstocks with similar effects on the overall horticultural performance of 'Tahiti' lime tress (Fig. 2).

Table 2
Leaf colorimetric variables in 2008 and mean drought tolerance scores (2006–2008) of non-irrigated 'Tahiti' lime budded onto 12 rootstocks. Bebedouro, SP, Brazil.

Rootstock	Drought tolerance score*	Leaf colorimetric variables**		
		L^*	C^*	h°
'Cravo FCAV' 'Rangpur' lime	3.00 a	37.60 c	21.15 abc	122.87 ab
'Cravo Limeira' 'Rangpur' lime	2.98 a	36.36 c	19.51 c	124.30 ab
'Sunki' mandarin	2.23 ab	35.67 c	18.08 c	124.06 ab
'HRS 801' hybrid	2.25 ab	35.50 c	17.25 c	125.02 a
'HRS 827' hybrid	1.83 bc	36.65 bc	19.37 bc	123.84 ab
'Rubidoux' trifoliate	1.75 bc	38.98 abc	22.62 abc	122.30 ac
'Carrizo' citrange	1.70 bc	36.64 bc	18.80 bc	123.62 ab
'Orlando' tangelo	1.58 bc	38.33 abc	21.71 abc	122.12 abc
'Swingle' citrumelo	1.55 bc	36.91 bc	20.12 bc	123.17 ab
'Sun Chu Sha Kat' mandarin	1.55 bc	37.32 bc	20.52 bc	123.00 ab
'FCAV' trifoliate	1.43 bc	42.08 a	26.90 a	118.45 c
'Flying Dragon' trifoliate	1.05 c	39.94 ab	23.98 ab	120.61 bc
CV (%)	37.04	2.71	8.36	1.59
P-value	<0.0001	<0.0001	<0.0001	<0.0001

Scores 1, 2 and 3: low, medium and high canopy tolerance to drought, respectively. Means followed by different letters in columns are significantly different at the indicated *P*-value by *Friedman's test and **Fisher's test. Original data of L^* and C^* were transformed to follow a normal distribution.

Table 3
Export fruit percentage, packing carton fruit sizes and peel colorimetric variables for export 'Tahiti' limes budded onto 12 rootstocks. Bebedouro, SP, Brazil, March 2007.

Rootstock	Export fruit (%)	Packing carton size class (%)		Peel colorimetric variables[#]		
		Small size[*]	Large size[**]	L^*	C^*	h°
'Flying Dragon' trifoliate	51.1 a	5.6 c	60.0 a	42.78	33.28	118.64
'FCAV' trifoliate	43.1 ab	21.1 bc	36.7 bc	44.33	33.48	118.93
'Rubidoux' trifoliate	43.4 ab	15.6 bc	38.9 ab	42.91	32.51	120.08[¥]
'Sun Chu Sha Kat' mandarin	43.8 ab	16.7 bc	36.7 bc	42.49	32.05[¥]	120.15[¥]
'Carrizo' citrange	39.4 abc	20.0 bc	30.0 bcd	45.35[¥]	33.27	119.76[¥]
'HRS 801' hybrid	38.0 abc	22.2 bc	22.4 bcd	43.50	31.27[¥]	120.28[¥]
'Swingle' citrumelo	36.2 abcd	21.1 bc	31.1 bcd	44.04	32.17[¥]	119.49[¥]
'Sunki' mandarin	25.3 bcd	25.6 ab	27.8 bcd	41.50[¥]	32.39[¥]	119.42[¥]
'HRS 827' hybrid	22.7 bcd	32.2 ab	12.2 d	45.56[¥]	34.10	119.73[¥]
'Orlando' tangelo	28.5 abcd	31.1 ab	12.2 d	42.96	32.53	119.53[¥]
'Cravo FCAV' 'Rangpur' lime	14.9 cd	28.9 ab	10.0 d	41.41[¥]	32.96	118.98
'Cravo Limeira' 'Rangpur' lime	12.1 d	41.1 a	16.7 cd	43.90	33.72	118.53
CV (%)	25.79	25.80	26.24	53.85	56.74	54.49
P-value	0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

[#] Means followed by different letters in columns are significantly different at the indicated *P*-value (Friedman's test) by the Tukey-Kramer's test ($P < 0.05$).
[*] Carton size classes 64 and 60.
[**] Carton size classes 48 and 42.
[¥] Significantly different from 'Cravo Limeira' 'Rangpur' lime by the Dunnet's test ($P < 0.05$).

Table 4
Mean values of fruit quality variables of 'Tahiti' limes for the domestic market harvested during the first semester as influenced by 12 rootstocks. Bebedouro, SP, Brazil, February–March, 2006–2008.

Rootstock	Fruit weight (g)	Fruit diameter (cm)	Fruit height (cm)	*JC* (%)	*TSS* (°Brix)	*TA* (%)
'Flying Dragon' trifoliate	87.3 ab	5.32 ab	5.87	45.25	8.42 a	6.37 ab
'FCAV' trifoliate	87.1 ab	5.32 ab	5.90	45.25	8.21 ab	6.59 a
'Rubidoux' trifoliate	89.4 a	5.33 a	6.05	47.08	8.12 abc	6.26 b
'Sunki' mandarin	84.0 ab	5.21 ab	5.86	47.99	7.88 bcd	6.56 ab
'Swingle' citrumelo	86.3 ab	5.32 ab	5.88	46.19	7.96 bcd	6.44 ab
'HRS 801' hybrid	85.3 ab	5.24 ab	5.86	46.69	8.13 ab	6.27 ab
'Sun Chu Sha Kat' mandarin	85.8 ab	5.26 ab	5.92	45.48	8.10 abc	6.52 ab
'Carrizo' citrange	83.3 ab	5.32 ab	5.93	46.37	7.90 bcd	6.46 ab
'HRS 827' hybrid	84.8 ab	5.29 ab	5.86	47.43	7.86 bcd	6.58 ab
'Cravo FCAV' 'Rangpur' lime	84.8 ab	5.29 ab	5.81	47.34	7.78 cd	6.38 ab
'Cravo Limeira' 'Rangpur' lime	87.5 ab	5.33 a	5.84	47.05	7.76 cd	6.36 ab
'Orlando' tangelo	80.6 b	5.15 b	5.79	44.89	7.66 d	6.36 ab
CV (%)	4.72	1.72	2.28	4.68	2.33	32.55
P-value	0.0469	0.0210	0.1818	0.2493	<0.0001	0.0043

JC, juice content; *TSS*, total soluble solids; *TA*, titratable acidity. Means followed by different letters in columns significantly differ at the indicated *P*-value (Fisher's test) by the Tukey's test ($P < 0.05$).

The first cluster of rootstocks with similar effects was comprised of the 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' limes, the 'Orlando' tangelo, the 'Sunki' mandarin and the 'HRS 827' hybrid. Within this first cluster, both 'Rangpur' lime rootstocks had very similar effects on the overall 'Tahiti' lime performance. A second cluster of comparable rootstocks included the 'Carrizo' citrange, 'HRS 801', 'FCAV' trifoliate, 'Swingle' citrumelo, 'Sun Chu Sha Kat' mandarin and the 'Rubidoux' trifoliate. In this group, the 'HRS 801' and the 'FCAV' trifoliate rootstocks induced very similar effects to the 'Tahiti' scion. On the other hand, the 'Flying Dragon' trifoliate induced unique responses over 'Tahiti' lime trees, different from the other evaluated rootstocks.

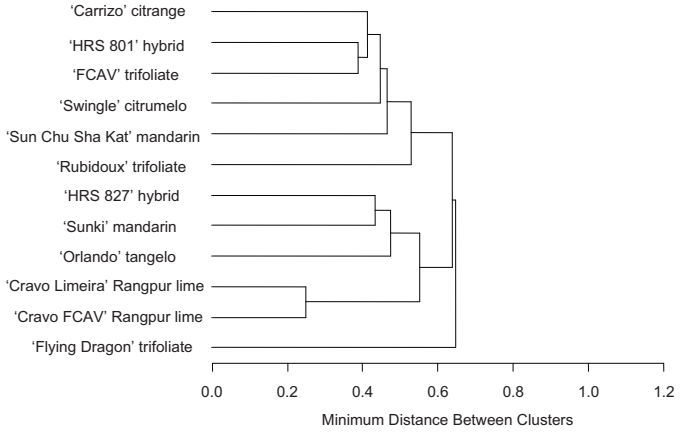

Fig. 2. Dendrogram of cluster analysis obtained by the single linkage method showing the similarity among groups of rootstocks based on their performance for 12 horticultural variables of 'Tahiti' lime budded onto 12 rootstocks.

4. Discussion

In Brazil, the 'Cravo Limeira' 'Rangpur' lime is the most widely used rootstock for rain-fed citrus production and particularly for the 'Tahiti' lime scion, because it induces good agronomical performance under non-irrigated conditions (Stenzel and Neves, 2004). In this study, 'Tahiti' lime scion attained highest cumulative yields and larger fruit size during the main harvest period in the first semester when grafted onto the 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' limes, thus confirming the expected effects of these rootstocks. Nevertheless, they both induced the lowest percentages of fruit classified for export, probably as a consequence of earlier fruit bearing induced by 'Rangpur' lime rootstocks, causing earlier color break and peel yellowing of 'Tahiti' limes that make them unsuitable for export.

'Tahiti' lime trees grafted onto the 'Flying Dragon' trifoliate were early bearers, as they produced the highest percentage of cumulative yield in the second commercial crop, followed by plants grafted onto 'Sunki' mandarin, 'Rubidoux' trifoliate and 'Swingle' citrumelo. Early bearing of citrus plants grafted onto the 'Flying Dragon' rootstock was also observed on non-irrigated 'Okitsu' Satsuma mandarin (Cantuarias-Avilés et al., 2010) and 'Folha Murcha' sweet orange trees (Cantuarias-Avilés et al., 2011).

Comparison of cumulative yield separately harvested during the first and the second semester until the seventh year after planting indicated that the plants grafted onto 'Rubidoux' trifoliate and 'Sun Chu Sha Kat' mandarin had an even harvest distribution throughout the year, while all the other rootstocks induced larger crop during the first semester of the year.

The rootstocks 'Orlando' tangelo, 'Cravo FCAV' 'Rangpur' lime, 'HRS 801' and 'Carrizo' citrange induced larger canopy volumes to 'Tahiti' lime trees. These results confirmed those observed by Espinoza-Nuñez et al. (2011) for non-irrigated 'Tahiti' lime trees. The invigorating effect of those rootstocks restricts their use for high density groves.

Although the 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' lime rootstocks induced highest cumulative yield and larger fruit size to the 'Tahiti' scion, they were less efficient in terms of fruit load produced per unit canopy volume. On the other hand, the trifoliate rootstocks induced high yield efficiency and fruit quality to 'Tahiti' lime trees, particularly the 'Flying Dragon' trifoliate. The same effects have been previously attributed to this rootstock for several non-irrigated citrus scions, such as 'Tahiti' lime (Espinoza-Nuñez et al., 2011), 'Okitsu' Satsuma mandarin (Cantuarias-Avilés et al., 2010), 'Oneco' mandarin (Gonzatto et al., 2011) and 'Folha Murcha' sweet orange (Cantuarias-Avilés et al., 2011). High yield efficiency and low vigor conferred by the 'Flying Dragon' rootstock are important attributes for high-density plantings. However, its low drought tolerance restricts its use to the availability of supplementary irrigation (Figueiredo et al., 2003; Stuchi et al., 2003).

Under rain-fed conditions, the high drought susceptibility of 'Tahiti' trees grafted onto the 'Flying Dragon' observed at the end of the winter dry season would induce delayed vegetative flushing in the plants. In the 'Folha Murcha' sweet orange, delayed shoot flushing of plants grafted onto the 'Flying Dragon' rootstock may cause the new vegetation to skip the periods of high pest infestations in the early and mid-spring periods, thus explaining the lower disease incidence observed on the plants grafted onto this rootstock (Cantuarias-Avilés et al., 2011). This effect may be another important advantage supporting the use of the 'Flying Dragon' trifoliate as rootstock on citrus groves under high disease and pest pressure conditions, as those currently affecting the main growing regions in São Paulo State.

Moderate drought tolerance observed on 'Tahiti' plants grafted onto 'Orlando' tangelo and 'Swingle' citrumelo confirms previous results (Figueiredo et al., 2003). High drought tolerance of 'Tahiti' lime trees grafted onto the 'Cravo FCAV' and 'Cravo Limeira' 'Rangpur' limes, as well as onto the 'Sunki' mandarin and the 'HRS 801' hybrid, was previously reported on non-irrigated 'Folha Murcha' sweet oranges grafted onto these rootstocks (Cantuarias-Avilés et al., 2011).

In this study, colorimetric evaluation of leaves in the period of maximum drought stress was a useful tool for determining differences on drought tolerance among rootstocks. Unlike the subjective evaluation based on scores of visual canopy water stress symptoms, leaf colorimetric measurements allow direct and objective determination of plant water stress for various citrus scions (Deidda et al., 1996; Cantuarias-Avilés et al., 2011).

Percentage fruit yield classified for domestic and foreign markets during the main harvest of 2007 was utilized to identify the most promising rootstocks for fruit production oriented to each particular target market. 'Tahiti' lime plants grafted onto the 'Flying Dragon' and 'FCAV' trifoliates yielded higher percentages of export fruit, with larger size and adequate peel color. On the other hand, 'Tahiti' lime plants grafted onto the 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' limes produced higher percentages of fruit suitable for domestic market, but induced small fruit size for export market. These results indicate that the use of 'Flying Dragon' and 'FCAV' trifoliates as rootstocks for 'Tahiti' lime production during the first semester may enhance export fruit yield and crop profitability even under non-irrigated conditions.

A multivariate evaluation of the overall performance of twelve rootstocks for 7 year-old 'Tahiti' lime trees under rain-fed conditions allowed to identify the 'Cravo Limeira' and 'Cravo FCAV 'Rangpur' limes as promising rootstocks for fruit production oriented to the domestic market during the first semester. Despite of its high susceptibility to drought stress under non-irrigated conditions, the 'Flying Dragon' trifoliate performed remarkably as rootstock for the 'Tahiti' lime scion, producing dwarf trees with highest yield efficiency on a canopy volume basis and inducing early bearing and large-sized fruits of good quality for foreign market. Contrarily, the 'Orlando' tangelo performed poorly as a rootstock for 'Tahiti' lime trees, inducing low cumulative yield, large plant size, lowest yield efficiency, and low fruit quality. Similar effects of the 'Orlando' tangelo had been previously reported when utilized as rootstock for non-irrigated 'Fallglo', 'Sunburst' and 'Okitsu' Satsuma mandarins (Mourão Filho et al., 2007; Cantuarias-Avilés et al., 2010).

5. Conclusions

The 'Cravo Limeira' and 'Cravo FCAV' 'Rangpur' lime rootstocks induced larger cumulative yield, conferring the highest drought tolerance to 'Tahiti' lime trees under rain-fed conditions.

The 'Flying Dragon' and 'FCAV' trifoliate rootstocks induced early bearing and higher yield efficiency to non-irrigated 'Tahiti' lime trees, with larger production of high quality fruits. Smaller sized 'Tahiti' plants grafted onto the 'Flying Dragon' trifoliate rootstock are better suited to high-density plantings, even though their low drought tolerance restricts their use to the availability of supplementary irrigation.

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References

ABPEL, Associação Brasileira de Produtores e Exportadores de Limão, 2011. http://www.abpel.org.br (20.05.11).

Cantuarias-Avilés, T., Mourão Filho, F.A.A., Stuchi, E.S., Silva, S.R., Espinoza-Nuñez, E., 2010. Tree performance and fruit yield and quality of 'Okitsu' Satsuma mandarin grafted on 12 rootstocks. Sci. Hortic. 123, 318–322.

Cantuarias-Avilés, T., Mourão Filho, F.A.A., Stuchi, E.S., Silva, S.R., Espinoza-Nuñez, E., 2011. Horticultural performance of 'Folha Murcha' sweet orange onto twelve rootstocks. Sci. Hortic. 129, 259–265.
Deidda, P., Spano, D., Arca, B., Ventura, A., Duce, P., 1996. Colorimetric analysis for determining leaf water status in *Citrus*. In: Proceedings 8th International Citrus Congress, pp. 1065–1068.
EMBRAPA, 1999. Centro Nacional de Pesquisa de Solos. Sistema Brasileiro de Classificação de Solos, Rio de Janeiro, p. 412.
Espinoza-Nuñez, E., Mourão Filho, F.A.A., Stuchi, E.S., Cantuarias-Avilés, T., Santos Dias, C.T., 2011. Performance of 'Tahiti' lime on twelve rootstocks under irrigated and non-irrigated conditions. Sci. Hortic. 129, 227–231.
Figueiredo, J.O., Laranjeira, F.F., Sobrinho, J.T., Pio, R.M., 2003. Rootstocks for 'Tahiti' lime in São Paulo State, Brazil. In: Proceedings of the International Society of Citriculture, vol. 1, pp. 487–489.
Gonzatto, M.P., Kovaleski, A.P., Brugnara, E.C., Weiler, R.L., Sartori, I.A., Lima, J.G., Bender, R.J., Schwarz, S.F., 2011. Performance of 'Oneco' mandarin on six rootstocks in South Brazil. Pesqui. Agropecu. Bras. 46, 406–411.
IBRAF, Instituto Brasileiro de Frutas, 2011. http://www.ibraf.org.br (20.05.11).
Manly, B.F.J., 2005. Multivariate Statistical Methods: A Primer, third ed. Chapman & Hall, London, p. 214.
Mattos Júnior, D., De Negri, J.D., Pio, R.M., Pompeu Júnior, J., 2005. Citros. Instituto Agronômico/FUNDAG, Campinas, p. 929.
McGuire, R.G., 1992. Reporting of objective color measurements. HortScience 27, 1254–1255.
Mourão Filho, F.A.A., Espinoza-Núñez, E., Stuchi, E.S., Ortega, E.M.M., 2007. Plant growth, yield, and fruit quality of 'Fallglo' and 'Sunburst' mandarins on four rootstocks. Sci. Hortic. 114, 45–49.
Silva, P.R., Francisco, V.L.F.S., Baptistella, C.S.L., 2008. Caracterização da cultura do limão no Estado de São Paulo 2001–2007. Inform. Econ. 38, 24–31.
Stenzel, N.M.C., Neves, C.S.V.J., 2004. Rootstocks for 'Tahiti' lime. Sci. Agric. 61, 151–155.
Stuchi, E.S., Donadio, L.C., Sempionato, O.R., 2000. Tolerância à seca da laranjeira 'Folha Murcha' em 10 porta-enxertos. Rev. Bras. Fruticult. 22, 454–457.
Stuchi, E.S., Donadio, L.C., Sempionato, O.R., 2003. Performance of Tahiti lime on *Poncirus trifoliata* var. *montrosa* Flying Dragon on four densities. Fruits 58, 13–17.
Zekri, M., 2000. Citrus rootstocks affect scion nutrition, fruit quality, growth, yield and economical return. Fruits 55, 231–239.