



# Article Rootstocks Genotypes Impact on Tree Development and Industrial Properties of 'Valencia' Sweet Orange Juice

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Abstract: The low diversification of rootstock genotypes in orchards limits the expansion of the citrus industry, restricting increases in productivity and cost-saving via phytosanitary treatments and other horticultural practices. Therefore, the aim of this study was to assess the impact of rootstock genotypes on tree development and industrial properties of 'Valencia' sweet orange juice (Citrus sinensis). Twenty rootstock genotypes were evaluated by measuring tree growth and industrial properties of orange juices, including 'Trifoliata' hybrids with tangerine (citrandarins) and grapefruit (citrumelos), as well 'Rangpur' lime and other potential rootstocks. The experimental orchard was planted in Rancho Alegre, PR, Brazil, under clay soil and subtropical rainfed conditions. A randomized block design with four replicates was used. Trees grown on IPEACS-239 and IPEACS-256 citrandarins, and on 'US-802' pummelo hybrid had low vigor, high production efficiency and high industrial properties of orange juice, and are therefore potential alternatives for high-density plantings. The F.80–3 and F.80–5 citrumelos also had good dwarfing potential and high production efficiency, but lower industrial properties of juice compared to the other 'Trifoliata' hybrid rootstocks. Trees grown on 'US-812' citrandarin rootstock had low vigor, good productive performance, accumulated production and production efficiency similar to 'Rangpur' lime, and high industrial properties of juices. Although the 'Rangpur' lime and the 'Florida' rough lemon allowed high yields, the trees are very vigorous, with low-quality fruits. A Quick Reference Chart was created to provide practical and objective identification of the best rootstock alternatives for 'Valencia' orange trees in terms of tree development and industrial properties of juices.

**Keywords:** *Citrus sinensis* (L.) Osb; rootstock genotypes; yield performance; dwarfism; tree growth; orange juice

# 1. Introduction

Citrus is the most economically important fruit crop in the world, and commercial sweet orange orchards consist of two-part trees that incorporate favorable attributes of the scion and rootstock. The selection of rootstock is the major factor to prepare plant material, and it should be carefully considered because citrus rootstocks can influence important aspects of the scion, including tree growth development, production, industrial properties of juices, disease resistance, as well as abiotic aspects such as cold and drought tolerance [1–4]. In other words, without rootstocks, own-rooted orange trees may have a poor development or even die in a short period of time. However, only few rootstock varieties are used in citrus cultivation in many countries, and this poses a health-risk for trees, and often limits the performance of scion varieties and orchard yield [5–7].

In Brazil, the largest citrus producer in the world, the main citrus rootstocks are 'Rangpur' lime (*Citrus limonia*) and 'Swingle' citrumelo (*Citrus paradisi* × *Poncirus trifoliata*).



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Orange trees grown on the 'Rangpur' lime have good yield, early production, drought tolerance and good vigor in the nursery, which explains its frequent use. However, it is susceptible to *Citrus exocortis viroid* (CEVd), *Citrus cachexia viroid* (CCaV) and to Citrus sudden death (CSDaV), and promotes fruits of lower quality than other rootstocks such as the 'Swingle' citrumelo and 'Trifoliata' (*P. trifoliata*) [8–12]. 'Swingle' citrumelo is a suitable rootstock for most soils other than heavy clay or highly calcareous soils. It is tolerant to *Citrus tristeza virus* (CTV) and CEVd, CCaV, citrus blight and citrus sudden death, and is resistant to *Phytophthora* gummosis (*Phytophthora* spp.) and citrus nematodes (*Tylenchulus semipenetrans*) [13–15]. However, all commercial scions and rootstocks used in Brazil, including the 'Rangpur' lime and 'Swingle' citrumelo, are sensitive to Huanglongbing (HLB), the current predominant disease in the citrus industry [2,16]. Therefore, it is extremely important to select different genotypes that provide agronomic characteristics equivalent or superior to those attributed to the main rootstocks in use.

The diversification of rootstocks should also prioritize those that provide better fruit quality to meet the demands of the juice industry, which requires fruits with a higher content of soluble solids, and thus, better industrial properties of orange juice [2,17]. In particular, the 'Valencia' orange tree (*Citrus sinensis*), is one of the main commercial oranges produced globally, with fruits used for processing, mainly in the form of frozen concentrated juice (FCOJ) and not concentrated juice (NFC) [18]. The production of raw materials must be of high quality to maintain the market competitiveness of this sector, and the fruit quality attributes are determined by their industrial properties, which in turn vary during the ripening period [19].

In terms of production, rootstock genotypes with dwarfing potential are good options for high-density planting, as they favor an increase in yield and contribute to reducing production costs [20,21]. Moreover, high-density planting favors the management of HLB, by compensating for the reduction in yield due to the mandatory removal of diseased trees, and due to tree removal to control the insects that spread the disease between trees [22–25].

More recently, two groups of 'Trifoliata' hybrid rootstocks in particular have emerged as important alternatives: citrandarins (*Citrus reticulata*  $\times$  *P. trifoliata*) and citrumelos (*C. paradisi*  $\times$  *P. trifoliata*). These rootstocks can promote high yields, higher quality fruit and dwarf trees that facilitate high-density planting, and promote resistance to some diseases [10,24,26–31]. Therefore, due to the lack of information about the performance of these rootstocks, our objective was to conduct a trial to evaluate various citrandarin and citrumelo rootstocks for 'Valencia' sweet orange grown under subtropical conditions, as well 'Rangpur' lime and other potential rootstocks. Special focus was given to identify the rootstocks that reduced tree size and improved chemical and industrial properties of orange juice.

# 2. Materials and Methods

#### 2.1. Location, Treatments and Experimental Design

The trial was conducted in a commercial citrus orchard in Rancho Alegre city (23°03'15" S, 50°55'50" W, elevation 380 m a.s.l.), northern Paraná State, Brazil. The climate of the region is subtropical humid (Cfa; Köppen classification), with hot summers, infrequent frosts, rains predominantly occurring in the summer months and without a well-defined dry season [32]. The annual mean rainfall is 1300 mm, with a mean temperature of 22.1 °C, relative humidity of 75–80% [33] and the predominant soil type is eutrophic red latosol.

## 2.2. Origin of Rootstocks Genotypes

The treatments included 20 rootstock genotypes, including hybrids of 'Trifoliata' (*P. trifoliata*), namely, citrandarins and citrumelos, as well 'Rangpur' lime and other potential rootstocks. The origin of the rootstock genotypes is presented in Table 1.

| Rootstocks                               | Parentage/Species  | Accession Codes/Origin<br>(CCSM/IAC BAG <sup>1</sup> and Certified Citrus Nursery <sup>2</sup> ) |  |  |  |  |
|--|--|--|--|--|--|--|
| 'US-852' citrandarin                     | Citrus reticulata 'Changsha' $	imes$ Poncirus trifoliata 'English Large' | 1454   |  |  |  |  |
| 'US-801' citrandarin                     | C. reticulata 'Changsha' $\times$ P. trifoliata 'English Small'          | 1710   |  |  |  |  |
| 'US-812' citrandarin                     | C. reticulata 'Sunki' $\times$ P. trifoliata 'Benecke'                   | 1697   |  |  |  |  |
| IPEACS-239 citrandarin                   | C. reticulata 'Cleopatra' $\times$ P. trifoliata 'Rubidoux'              | 1600   |  |  |  |  |
| IPEACS-256 citrandarin                   | C. reticulata 'Cleopatra' $\times$ P. trifoliata 'English'               | 1483   |  |  |  |  |
| IPEACS-264 citrandarin                   | C. reticulata 'Sunki' $\times$ P. trifoliata 'English'                   | 1628   |  |  |  |  |
| F.80–3 citrumelo                         | Citrus paradisi $\times$ P. trifoliata                                   | 1460   |  |  |  |  |
| F.80–5 citrumelo                         | C. paradisi $\times$ P. trifoliata                                       | 1457   |  |  |  |  |
| F.80–6 citrumelo                         | C. paradisi × P. trifoliata  | 1456   |  |  |  |  |
| F.80–7 citrumelo                         | C. paradisi × P. trifoliata  | 1458   |  |  |  |  |
| F.80–8 citrumelo                         | C. paradisi $\times$ P. trifoliata                                       | 1459   |  |  |  |  |
| 'W-2' citrumelo                          | C. paradisi $\times$ P. trifoliata                                       | 1455   |  |  |  |  |
| 'Swingle' citrumelo                      | C. paradisi $\times$ P. trifoliata                                       | 401  |  |  |  |  |
| 'US-802' pummelo hybrid                  | Citrus grandis 'Siamese' × P. trifoliata 'Gotha Road'                    | 1690   |  |  |  |  |
| 'Murcott' tangor $\times$ 'Trifoliata'–9 | (C. reticulata $\times$ C. sinensis) $\times$ P. trifoliata              | 1470   |  |  |  |  |
| 'Trifoliata'                             | P. trifoliata  | Certified Citrus Nursery   |  |  |  |  |
| 'Flying Dragon'                          | P. trifoliata 'Flying Dragon'  | Certified Citrus Nursery   |  |  |  |  |
| 'Rangpur' lime                           | Citrus limonia Osb.  | Certified Citrus Nursery   |  |  |  |  |
| 'Florida' rough lemon                    | Citrus jambhiri Lush.  | Certified Citrus Nursery   |  |  |  |  |
| 'Sunki' tangerine                        | C. reticulata 'Sunki'  | Certified Citrus Nursery   |  |  |  |  |

Table 1. Common names, parentage (species names), access code or origin of 20 rootstocks evaluated for 'Valencia' sweet orange.

<sup>1</sup> CCSM/IAC BAG: Germplasm Bank—Centro de Citricultura Sylvio Moreira, Instituto Agronômico de Campinas, IAC, Brazil. <sup>2</sup> Pratinha Certified Citrus Nursery (Genotypes from Germplasm Bank–IDR–Instituto Agronômico do Paraná, IAPAR), Brazil.

Field-ready grafted 'Valencia' orange trees clone IAC (*C. sinensis*) were planted in April 2013 in a  $6.0 \times 2.5$  m spacing, at 666.7 trees ha<sup>-1</sup>, under rainfed conditions. A randomized complete block design was used, with 20 treatments and four replicates, and each plot contained four trees.

## 2.3. Agricultural Practices

Liming was performed before planting the trees to neutralize soil acidity, provided that the initial pH value was 5.7. Powdered limestone was applied throughout the area in 2 m wide planting range around the nursery trees, at a depth of 20 cm. The pits, prepared in a deep groove, received 500 g of single superphosphate per nursery tree, 5 L of organic fertilizer and 200 g of limestone per linear meter [34]. Phytosanitary control was performed by monitoring pests and diseases, controlling them when they reached damage levels and weeds were controlled with herbicides within rows, and mowing between rows. Other agricultural practices were developed following the technical recommendations for the region [35]. No pruning was performed in the experimental orchard during the evaluated period.

## 2.4. Evaluations

#### 2.4.1. Tree Growth Development

The growth development of 'Valencia' orange trees was assessed by measuring tree height (the distance from base to top of the tree) and canopy diameter, which were used to calculate canopy volume (*V*) using the following equation [36]:

$$V\left(\mathbf{m}^{3}\right) = \frac{2}{3} \times \left(\pi R^{2} H\right)$$

where *R* is the canopy radius and *H* is the tree height. Two orthogonal measurements were made in the middle of the tree, using a graduated scale, to determine the diameter and radius of the tree canopy. Tree growth was assessed in autumn (May and June) 2017, 2018 and 2019.

# 2.4.2. Orange Production

To determine the production of each scion/rootstock combination (in kg fruit per tree), the fruits from each plot were weighed digitally at harvest in October 2017, 2018 and 2019. Accumulated fruit production per tree was the sum of production for the three harvest

seasons. Based on these results, the production efficiency (*PE*) for the harvest seasons was calculated as follows:

$$PE(kg m^{-3}) = \frac{Fruit \text{ production } (kg/tree)}{Canopy \text{ volume } (m^3)}$$

2.4.3. Chemical and Industrial Properties of Orange Juices

The analyses of the impact of rootstocks on the main chemical and industrial properties of 'Valencia' orange juices were performed using samples of 15 fruits per plot, sampled at random from the outside of the trees, at a height of 1.0–2.0 m. Fruit samples were weighed and juiced using an FMC citrus juice extractor, and juice quality was analyzed using the standard laboratory methods [37] at the Cooperativa Integrada Citrus Processing Plant, Uraí, PR, Brazil. The assessed industrial properties of orange juice are described below.

Soluble solid (SS) content of each sample was determined by direct reading, using a PAL– $\alpha^{\text{(B)}}$  digital refractometer (Atago, Tokyo, Japan) with automatic temperature compensation, and the results were expressed in °Brix. Titratable acidity (TA) was determined by titrating 25 mL of juice with 0.1 N sodium hydroxide (NaOH) solution at pH 8.2, and is expressed as the percentage of citric acid. The maturation index (SS/TA), was used to evaluate fruit maturity.

Juice yield (*JY*, %) was determined using the following equation:

$$JY(\%) = \frac{\text{Juice mass } (g)}{\text{Fruit mass } (g)} \times 100.$$

The technological index (*TI*), the quantity of juice *SS* in 40.8 kg of fruit expressed as kg *SS* per 40.8 kg box was calculated using the following equation:

$$I = \frac{[JY (\%) \times SS (°Brix) \times 40.8 \text{ kg (weight of a standard orange box)}]}{10,000}$$

The industrial yield (IY), the number of orange boxes required to obtain 1 ton of frozen concentrated orange juice (66 °Brix), was calculated using the following equation:

$$IY = \frac{660}{TI}$$

where 660 represents the quantity (660 kg) of SS in 1 ton of frozen concentrated juice (66 °Brix).

## 2.5. Statistical Analyses

Data on vegetative development, production, chemical and industrial properties of orange juices were subjected to one-way ANOVA with the rootstock as variability factor, complemented by the Scott–Knott test (p < 0.05 significance threshold) for all data, using Sisvar<sup>®</sup> software [38]. Principal component analysis was performed to explain the relationships between rootstocks and the evaluated variables, which were later grouped using agglomerative hierarchical clustering, both using R software and the FactorMiner package.

Using cluster analysis of production per tree, production efficiency, technological index and industrial yield for the 2017, 2018 and 2019 harvest seasons, and canopy volume for the 2019 harvest season, a Quick Reference Chart was created based on Castle et al. [26]. This chart accounts for the influence of each rootstock on the characteristics of the 'Valencia' orange tree, such as tree development and industrial properties of orange juice.

# 3. Results

# 3.1. Tree Growth Development

'Valencia' orange trees grown on 'Flying Dragon' trifoliate, F.80–3 and F.80–5 citrumelo had lower canopy height than those grown on 'Florida' rough lemon and the 'Rangpur' lime rootstocks. All of the scion/rootstock combinations had canopy height <3.0 m, except for those using the IPEACS–264 citrandarin, F.80–8, F.80–7 and 'W–2' citrumelos, 'Rangpur' lime and 'Florida' rough lemon rootstocks. Some rootstock genotypes have dwarfing potential [39], and we observed this with heights  $\leq$ 2.5 m for the IPEACS–256 (2.5 m) and IPEACS–239 citrandarins (2.4 m), F.80–3 (2.3 m) and F.80–5 (2.2 m) citrumelos and 'Flying Dragon' trifoliate (2.0 m) (Table 2).

Table 2. Tree growth development of 'Valencia' sweet orange trees grafted on 20 rootstocks.

| Bootstocks                               | Canopy Height (m) |        |        | Canopy Diameter (m) |        |       | Canopy Volume (m <sup>3</sup> ) |        |        |
|--|-------------------|--------|--------|---------------------|--------|-------|---------------------------------|--------|--------|
| ROUSIOCKS                                | 2017              | 2018   | 2019   | 2017                | 2018   | 2019  | 2017                            | 2018   | 2019   |
| 'US-852' citrandarin                     | 2.4 b             | 2.5 e  | 2.8 c  | 2.2 с               | 2.4 b  | 2.5 с | 6.0 d                           | 7.8 c  | 9.1 d  |
| 'US-801' citrandarin                     | 2.5 b             | 2.8 d  | 2.9 с  | 2.3 b               | 2.5 a  | 2.6 b | 7.0 c                           | 8.7 b  | 10.7 c |
| 'US-812' citrandarin                     | 2.5 b             | 2.3 f  | 2.7 d  | 2.3 b               | 2.4 a  | 2.5 c | 6.8 c                           | 7.2 с  | 8.7 d  |
| IPEACS-239 citrandarin                   | 2.3 c             | 2.1 g  | 2.4 d  | 2.2 c               | 2.3 b  | 2.3 d | 5.6 d                           | 6.0 d  | 6.6 e  |
| IPEACS-256 citrandarin                   | 2.2 c             | 2.2 g  | 2.5 d  | 2.2 c               | 2.4 a  | 2.4 c | 5.6 d                           | 6.5 d  | 7.2 e  |
| IPEACS-264 citrandarin                   | 2.7 b             | 3.0 c  | 3.4 b  | 2.3 b               | 2.5 a  | 2.9 a | 7.4 c                           | 9.6 b  | 14.8 b |
| F.80–3 citrumelo                         | 2.0 d             | 2.1 g  | 2.3 e  | 2.0 d               | 2.3 b  | 2.2 d | 4.3 e                           | 5.9 d  | 6.0 e  |
| F.80–5 citrumelo                         | 1.8 e             | 2.0 g  | 2.2 e  | 1.9 e               | 2.1 c  | 2.2 d | 3.3 f                           | 4.9 e  | 5.7 e  |
| F.80–6 citrumelo                         | 2.3 c             | 2.5 e  | 2.7 d  | 2.1 c               | 2.4 a  | 2.5 c | 5.6 d                           | 7.6 c  | 9.0 d  |
| F.80–7 citrumelo                         | 2.5 b             | 2.9 d  | 3.0 c  | 2.3 c               | 2.4 a  | 2.7 b | 6.7 c                           | 8.8 b  | 11.6 c |
| F.80–8 citrumelo                         | 2.5 b             | 3.0 c  | 3.1 c  | 2.2 c               | 2.5 a  | 2.8 b | 6.5 c                           | 9.4 b  | 12.2 c |
| 'W–2' citrumelo                          | 2.5 b             | 3.0 c  | 3.1 c  | 2.2 c               | 2.4 a  | 2.7 b | 6.3 c                           | 9.3 b  | 11.7 c |
| 'Swingle' citrumelo                      | 2.5 b             | 2.9 d  | 2.9 с  | 2.3 b               | 2.5 a  | 2.6 b | 6.7 c                           | 9.0 b  | 10.0 d |
| 'US-802' pummelo hybrid                  | 2.3 c             | 2.3 f  | 2.6 d  | 2.1 c               | 2.3 b  | 2.3 c | 5.4 d                           | 6.8 d  | 7.3 e  |
| 'Murcott' tangor $\times$ 'Trifoliata'–9 | 2.4 b             | 2.4 e  | 2.7 d  | 2.2 c               | 2.4 a  | 2.5 c | 6.1 d                           | 7.5 c  | 8.4 d  |
| 'Trifoliata'                             | 2.2 c             | 2.3 f  | 2.6 d  | 2.1 c               | 2.3 b  | 2.4 c | 5.4 d                           | 6.5 d  | 7.9 d  |
| 'Flying Dragon'                          | 1.8 e             | 1.8 h  | 2.0 e  | 1.8 e               | 2.0 d  | 2.1 d | 3.2 f                           | 3.6 f  | 4.7 e  |
| 'Rangpur' lime                           | 2.9 a             | 3.2 b  | 3.4 b  | 2.4 b               | 2.5 a  | 2.7 b | 8.2 b                           | 10.2 a | 13.2 b |
| 'Florida' rough lemon                    | 3.0 a             | 3.5 a  | 3.7 a  | 2.5 a               | 2.5 a  | 3.1 a | 9.6 a                           | 11.3 a | 18.9 a |
| 'Sunki' tangerine                        | 2.6 b             | 2.7 d  | 2.9 c  | 2.5 a               | 2.5 a  | 2.8 b | 8.2 b                           | 8.9 b  | 11.7 c |
| F–value                                  | 25.6 *            | 31.6 * | 20.1 * | 12.7 *              | 11.0 * | 8.5 * | 25.1 *                          | 25.8 * | 19.3 * |

Means within columns followed by the same letter do not differ statistically by one-way ANOVA with the rootstock as variability factor (Scott–Knott test, p < 0.05). \* p < 0.01.

Trees grown on 'Flying Dragon' trifoliate and on F.80–5 and F.80–3 citrumelo rootstocks had narrower canopy diameter (mean diameter 1.8–2.2 m for all seasons) than those on 'Florida' rough lemon (mean diameter 2.5–3.1 m). Trees grown on IPEACS–239 citrandarin rootstock had narrower canopy diameter than those grown on the other citrandarin rootstocks; those grown on F.80–3, F.80–5 and F.80–6 citrumelo rootstocks had narrower canopy diameter than those on the other citrumelo rootstocks (Table 2).

Trees on F.80–3 and F.80–5 citrumelo and 'Flying Dragon' trifoliate rootstocks had the lowest values in canopy volume (means of 6.0 m<sup>3</sup>, 5.7 m<sup>3</sup> and 4.7 m<sup>3</sup>, respectively, for the last season). Those on F.80–6 citrumelo, IPEACS–256 and IPEACS–239 citrandarins and 'US–802' pummelo hybrid had intermediate canopy volumes (6.6–9.0 m<sup>3</sup>). Conversely, in all seasons, canopy volume was highest for trees grown on 'Florida' rough lemon rootstock, followed by 'Rangpur' lime, 'Sunki' tangerine, F.80–7, F.80–8 and 'W–2' citrumelos and IPEACS–264 and 'US–801' citrandarins, with means of 10.7–18.9 m<sup>3</sup> (Table 2).

Due to the integrated management for mitigating HLB in the experimental location, such as monitoring and chemical control of psyllid, local and regional removal of symptomatic trees, release of psyllid parasitoids, among others, the disease incidence was low during the assessments and evenly distributed in the area, i.e., all rootstock genotype seemed to have the same behavior regarding this characteristic.

### 3.2. Orange Production

In the first harvest season (2017), trees grown on 'Rangpur' lime, 'US–812' citrandarin and 'W–2' citrumelo rootstocks had higher production, averaging 101.1, 91.4 and 89.0 kg/tree, respectively. The least productive rootstocks were F.80–3, F.80–5 and F.80–6 citrumelos, 'US–802' pummelo hybrid, 'Trifoliata' and 'Flying Dragon' trifoliate, with means ranging from 57.8 kg/tree to 26.9 kg/tree (Table 3).

| Rootstocks                               | Pro     | duction (kg/ | tree)   | Accumulated | Production Efficiency (kg m <sup>-3</sup> ) |       |        |  |
|--|---------|--------------|---------|-------------|---|-------|--------|--|
| ROUSIOCKS -                              | 2017    | 2018 2019    |         | Production  | 2017  | 2018  | 2019   |  |
| 'US–852' citrandarin                     | 55.2 c  | 46.9 b       | 65.7 b  | 167.8 b     | 9.0 b                                       | 6.1 b | 7.5 b  |  |
| 'US-801' citrandarin                     | 79.8 b  | 54.5 a       | 89.3 a  | 223.6 a     | 11.4 a                                      | 6.3 b | 8.5 b  |  |
| 'US-812' citrandarin                     | 89.0 a  | 69.4 a       | 66.1 b  | 224.4 a     | 13.2 a                                      | 9.7 a | 7.5 b  |  |
| IPEACS-239 citrandarin                   | 63.7 b  | 46.6 b       | 66.2 b  | 176.4 b     | 11.2 a                                      | 7.8 a | 10.3 a |  |
| IPEACS-256 citrandarin                   | 72.4 b  | 33.0 b       | 82.0 a  | 187.5 b     | 13.1 a                                      | 5.0 b | 11.4 a |  |
| IPEACS-264 citrandarin                   | 82.1 b  | 78.2 a       | 75.6 b  | 235.9 a     | 11.1 a                                      | 8.2 a | 5.1 b  |  |
| F.80–3 citrumelo                         | 57.8 c  | 42.0 b       | 72.0 b  | 171.8 b     | 13.5 a                                      | 6.9 a | 12.4 a |  |
| F.80–5 citrumelo                         | 46.8 c  | 39.4 b       | 61.6 b  | 147.8 b     | 14.1 a                                      | 8.3 a | 10.8 a |  |
| F.80–6 citrumelo                         | 48.5 c  | 54.6 a       | 73.4 b  | 176.5 b     | 9.0 b                                       | 7.3 a | 8.1 b  |  |
| F.80–7 citrumelo                         | 70.3 b  | 59.5 a       | 95.4 a  | 225.2 a     | 10.6 a                                      | 6.8 a | 8.7 b  |  |
| F.80–8 citrumelo                         | 72.3 b  | 60.3 a       | 95.2 a  | 227.8 a     | 11.1 a                                      | 6.6 a | 8.3 b  |  |
| 'W–2' citrumelo                          | 91.4 a  | 47.3 b       | 97.5 a  | 236.2 a     | 14.6 a                                      | 5.0 b | 8.4 b  |  |
| 'Swingle' citrumelo                      | 73.3 b  | 48.7 b       | 93.2 a  | 215.1 a     | 10.9 a                                      | 5.4 b | 9.6 a  |  |
| 'US-802' pummelo hybrid                  | 48.0 c  | 52.6 a       | 96.1 a  | 196.7 a     | 8.6 b                                       | 8.0 a | 13.3 a |  |
| 'Murcott' tangor $\times$ 'Trifoliata'–9 | 68.7 b  | 66.3 a       | 96.1 a  | 231.1 a     | 11.3 a                                      | 8.8 a | 11.6 a |  |
| 'Trifoliata'                             | 41.9 c  | 46.6 b       | 77.2 b  | 165.7 b     | 7.6 b                                       | 7.5 a | 9.9 a  |  |
| 'Flying Dragon'                          | 26.9 c  | 5.8 c        | 37.4 c  | 70.1 c      | 8.7 b                                       | 1.6 c | 8.0 b  |  |
| 'Rangpur' lime                           | 101.1 a | 62.0 a       | 100.8 a | 263.9 a     | 12.3 a                                      | 6.1 b | 7.5 b  |  |
| 'Florida' rough lemon                    | 82.2 b  | 73.2 a       | 98.7 a  | 254.1 a     | 8.6 b                                       | 6.5 a | 5.3 b  |  |
| 'Sunki' tangerine                        | 77.8 b  | 46.4 b       | 95.1 a  | 219.3 a     | 9.7 b                                       | 5.3 b | 8.2 b  |  |
| F–value                                  | 7.4 *   | 4.8 *        | 3.7 *   | 10.1 *      | 2.6 **                                      | 2.9 * | 4.3 *  |  |

Table 3. Production of 'Valencia' sweet orange trees grafted on 20 rootstocks.

Means within columns followed by the same letter do not differ statistically by one-way ANOVA with the rootstock as variability factor (Scott–Knott test, p < 0.05). \* p < 0.01. \*\* p < 0.05.

For the 2018 harvest season, trees on 'US–801', 'US–812' and IPEACS–264 citrandarins, F.80–6, F.80–7 and F.80–8 citrumelos, 'US–802' pummelo hybrid, 'Murcott' tangor  $\times$  'Trifoliata'–9, 'Rangpur' lime and 'Florida' rough lemon rootstocks had higher production, and those on F.80–3, F.80–5, 'W–2' and 'Swingle' citrumelo, 'US–852', IPEACS–256 and IPEACS–239 citrandarin, 'Flying Dragon' trifoliate, 'Trifoliata' and 'Sunki' tangerine rootstocks had lower production. For the 2019 harvest season, trees grown on at least eight of the 'Trifoliata' hybrid rootstocks had high production, with no statistical difference from that of those grown on the 'Rangpur' lime, 'Florida' rough lemon and 'Sunki' tangerine rootstocks (Table 3).

The accumulated production means were higher, at 196.7–263.9 kg/tree, for 'US–801', 'US–812' and IPEACS–264 citrandarin, F.80–7, F.80–8, 'W–2' and 'Swingle' citrumelo, 'US–802' pummelo hybrid and 'Murcott' tangor × 'Trifoliata'–9 rootstocks; their accumulated production did not differ statistically from that of the 'Rangpur' lime, 'Florida' rough lemon and 'Sunki' tangerine rootstocks. The F.80–3, F.80–5 and F.80–6 citrumelo rootstocks had lower mean accumulated production than the other citrumelos; 'US–852', IPEACS–256 and IPEACS–239 were the least productive among the citrandarin rootstocks (Table 3).

In the three harvest seasons assessed, trees on F.80–3 and F.80–5 citrumelo, IPEACS–239 citrandarin and 'Murcott' tangor  $\times$  'Trifoliata'–9 rootstock had better production efficiency, with means in the last harvest season of 10.8–12.4 kg m<sup>-3</sup>. The IPEACS–256, IPEACS–264 and 'US–812' citrandarins, F.80–7, F.80–8 and 'Swingle' citrumelos, 'US–802' pummelo hybrid and 'Trifoliata' rootstocks also had good production efficiency in at least two harvest seasons, with no difference among the other genotypes with the highest means. Conversely, 'Sunki' tangerine, 'US–852' and 'US–801' citrandarins, 'Flying Dragon' trifoliate, 'Rangpur' lime, 'Florida' rough lemon and 'W–2' and F.80–6 citrumelo rootstocks produced the lowest mean of production efficiency (Table 3).

#### 3.3. Chemical and Industrial Properties of Orange Juices

For the 2017 harvest season, trees grown on IPEACS–256 citrandarin and 'Flying Dragon' trifoliate rootstocks had fruits with the highest mean SS content (11.1 and 11.5 °Brix, respectively); the lowest mean SS content was obtained for 'Rangpur' lime (8.9 °Brix), 'Florida' rough lemon (8.6 °Brix) and F.80–7 citrumelo rootstocks (9.1 °Brix) (Table 4).

Table 4. Chemical properties of juices of 'Valencia' sweet orange trees grafted on 20 rootstocks.

| Rootstocks                        | Soluble Solids—SS (°Brix) |        |        | Titratable Acidity—TA (%) |        |                   | Maturation Index—MI<br>(SS/TA) |        |        |
|-----------------------------------|---------------------------|--------|--------|---------------------------|--------|-------------------|--------------------------------|--------|--------|
|                                   | 2017                      | 2018   | 2019   | 2017                      | 2018   | 2019              | 2017                           | 2018   | 2019   |
| 'US–852' citrandarin              | 10.4 b                    | 10.7 a | 12.4 a | 0.7 b                     | 0.7 b  | 0.5               | 14.4 a                         | 14.8 a | 24.4 a |
| 'US-801' citrandarin              | 9.7 c                     | 9.7 a  | 11.4 b | 0.8 b                     | 0.7 b  | 0.6               | 12.7 b                         | 14.1 a | 19.0 b |
| 'US-812' citrandarin              | 10.7 b                    | 10.7 a | 12.7 a | 0.8 b                     | 1.0 a  | 0.7               | 13.5 a                         | 11.3 b | 19.0 b |
| IPEACS-239 citrandarin            | 10.7 b                    | 10.9 a | 12.9 a | 0.8 b                     | 0.7 b  | 0.6               | 13.1 a                         | 16.3 a | 20.5 b |
| IPEACS-256 citrandarin            | 11.1 a                    | 11.4 a | 12.6 a | 0.8 b                     | 0.8 b  | 0.6               | 14.5 a                         | 14.4 a | 20.7 b |
| IPEACS-264 citrandarin            | 9.8 c                     | 9.6 a  | 12.2 a | 0.8 b                     | 0.7 b  | 0.7               | 13.0 a                         | 13.7 b | 19.7 b |
| F.80–3 citrumelo                  | 9.9 c                     | 9.3 b  | 11.3 b | 0.8 b                     | 0.7 b  | 0.6               | 12.8 b                         | 13.7 b | 18.3 b |
| F.80–5 citrumelo                  | 9.8 c                     | 10.3 a | 11.0 b | 0.8 b                     | 0.7 b  | 0.6               | 12.5 b                         | 14.9 a | 19.9 b |
| F.80–6 citrumelo                  | 9.9 c                     | 10.7 a | 11.9 b | 0.7 b                     | 0.7 b  | 0.6               | 13.6 a                         | 15.4 a | 18.6 b |
| F.80–7 citrumelo                  | 9.1 d                     | 9.1 b  | 11.1 b | 0.7 b                     | 0.7 b  | 0.6               | 12.3 b                         | 12.6 b | 18.0 b |
| F.80–8 citrumelo                  | 9.6 c                     | 10.0 a | 11.6 b | 0.8 b                     | 0.7 b  | 0.7               | 12.1 b                         | 14.5 a | 17.4 b |
| 'W–2' citrumelo                   | 9.6 c                     | 9.6 a  | 11.4 b | 0.8 b                     | 0.7 b  | 0.6               | 12.5 b                         | 13.8 b | 19.7 b |
| 'Swingle' citrumelo               | 10.0 c                    | 10.1 a | 12.7 a | 0.8 b                     | 0.8 b  | 0.7               | 13.3 a                         | 13.1 b | 17.9 b |
| 'US-802' pummelo hybrid           | 9.7 c                     | 9.8 a  | 11.7 b | 0.7 b                     | 0.7 b  | 0.6               | 13.1 a                         | 14.9 a | 20.1 b |
| 'Murcott' tangor × 'Trifoliata'–9 | 9.7 c                     | 9.7 a  | 12.6 a | 0.8 b                     | 0.7 b  | 0.7               | 12.3 b                         | 13.0 b | 18.3 b |
| 'Trifoliata'                      | 10.6 b                    | 10.6 a | 13.3 a | 0.8 b                     | 0.7 b  | 0.6               | 14.0 a                         | 15.9 a | 22.9 a |
| 'Flying Dragon'                   | 11.5 a                    | 11.1 a | 12.5 a | 1.0 a                     | 0.8 b  | 0.7               | 12.1 b                         | 15.1 a | 18.7 b |
| 'Rangpur' lime                    | 8.9 d                     | 9.2 b  | 11.7 b | 0.8 b                     | 0.7 b  | 0.7               | 11.5 b                         | 12.9 b | 17.4 b |
| 'Florida' rough lemon             | 8.6 d                     | 8.8 b  | 11.2 b | 0.8 b                     | 0.7 b  | 0.7               | 11.3 b                         | 12.9 b | 17.1 b |
| 'Sunki' tangerine                 | 9.8 c                     | 10.4 a | 12.2 a | 0.7 b                     | 0.7 b  | 0.6               | 13.6 a                         | 15.1 a | 20.4 b |
| F–value                           | 12.1 *                    | 8.7 *  | 2.7 *  | 3.9 *                     | 1.9 ** | 2.0 <sup>ns</sup> | 3.3 *                          | 3.0 *  | 2.4 *  |

Means within columns followed by the same letter do not differ statistically by one-way ANOVA with the rootstock as variability factor (Scott–Knott test, p < 0.05). \* p < 0.01. ns: not significant.

Those on 'Flying Dragon' trifoliate rootstock had fruits with higher TA than the other genotypes evaluated, and the highest maturation index—MI or *ratio* (SS/TA) was observed for fruits of trees grown on F.80–6 and 'Swingle' citrumelo, 'US–802' pummelo hybrid, 'Trifoliata', 'Sunki' tangerine and the other citrandarin rootstocks, except for 'US–801' (Table 4). The lowest means of industrial yield and technological index were observed for fruits of trees grown on 'US–801' citrandarin, F.80–5, F.80–7 and 'W–2' citrumelos, 'Rangpur' lime, and 'Florida' rough lemon (Table 5).

For the 2018 harvest season, trees on F.80–7 citrumelo, 'Rangpur' lime and 'Florida' rough lemon rootstocks had fruit with lower SS content. Higher fruit TA was observed when trees were grown on 'US–812' citrandarin rootstock. Mean MI was highest when trees were on 'US–852', 'US–801', IPEACS–256 and IPEACS–239 citrandarins, F.80–5, F.80–6 and F.80–8 citrumelos, 'US–802' pummelo hybrid, 'Trifoliata', 'Flying Dragon' trifoliate and 'Sunki' tangerine (Table 4). Trees grown on 'Florida' rough lemon had the highest mean industrial yield and lowest mean technological index (382.3 boxes of 40.8 kg each per ton of concentrated juice, and 1.7 kg of SS per 40.8 kg box, respectively). The order of the other rootstocks, ranked in terms of decreasing industrial yield, is as follows: 'Rangpur' lime, 'US–801' and IPEACS–264 citrandarins, F.80–3, F.80–7 and 'W–2' citrumelos and 'Murcott' tangor  $\times$  Trifoliata–9' (Table 5).

| Rootstocks                               | Juice Yield (%)   |                   |                   | Inc     | lustrial Yiel | d <sup>a</sup>    | Technological Index <sup>b</sup> |       |                   |
|--|-------------------|-------------------|-------------------|---------|---------------|-------------------|----------------------------------|-------|-------------------|
| Rootstocks                               | 2017              | 2018              | 2019              | 2017    | 2018          | 2019              | 2017                             | 2018  | 2019              |
| 'US–852' citrandarin                     | 60.9              | 53.7              | 58.8              | 258.1 b | 282.7 с       | 221.9             | 2.6 a                            | 2.3 a | 3.0               |
| 'US-801' citrandarin                     | 59.5              | 51.3              | 60.6              | 282.8 a | 326.2 b       | 236.6             | 2.4 b                            | 2.0 b | 2.8               |
| 'US-812' citrandarin                     | 61.4              | 55.0              | 63.9              | 251.6 b | 277.2 с       | 200.2             | 2.7 a                            | 2.4 a | 3.3               |
| IPEACS-239 citrandarin                   | 66.7              | 52.9              | 55.9              | 229.2 b | 280.4 c       | 225.4             | 2.9 a                            | 2.4 a | 2.9               |
| IPEACS-256 citrandarin                   | 61.3              | 51.6              | 56.9              | 239.4 b | 277.4 с       | 228.7             | 2.8 a                            | 2.4 a | 2.9               |
| IPEACS-264 citrandarin                   | 62.5              | 52.0              | 60.2              | 267.1 b | 326.9 b       | 222.8             | 2.5 a                            | 2.0 b | 3.0               |
| F.80–3 citrumelo                         | 62.2              | 50.9              | 59.0              | 265.7 b | 342.3 b       | 243.0             | 2.5 a                            | 1.9 b | 2.7               |
| F.80–5 citrumelo                         | 56.3              | 50.9              | 57.4              | 297.3 a | 309.8 c       | 257.6             | 2.2 b                            | 2.2 a | 2.6               |
| F.80–6 citrumelo                         | 61.3              | 53.0              | 58.9              | 268.8 b | 285.3 c       | 232.0             | 2.5 a                            | 2.3 a | 2.9               |
| F.80–7 citrumelo                         | 59.0              | 53.0              | 56.2              | 302.7 a | 338.4 b       | 262.8             | 2.2 b                            | 2.0 b | 2.6               |
| F.80–8 citrumelo                         | 63.0              | 52.7              | 59.5              | 268.3 b | 307.3 c       | 235.3             | 2.5 a                            | 2.2 a | 2.8               |
| 'W–2' citrumelo                          | 59.8              | 53.3              | 59.1              | 286.2 a | 315.4 b       | 243.1             | 2.3 b                            | 2.1 b | 2.7               |
| 'Swingle' citrumelo                      | 63.7              | 52.6              | 58.0              | 255.8 b | 303.8 c       | 221.3             | 2.6 a                            | 2.2 a | 3.0               |
| 'US–802' pummelo hybrid                  | 62.2              | 55.7              | 57.6              | 273.3 b | 295.8 с       | 243.0             | 2.5 a                            | 2.2 a | 2.7               |
| 'Murcott' tangor $\times$ 'Trifoliata'-9 | 63.0              | 51.2              | 57.2              | 264.1 b | 327.3 b       | 236.5             | 2.5 a                            | 2.0 b | 2.9               |
| 'Trifoliata'                             | 57.9              | 52.1              | 52.4              | 271.8 b | 305.6 c       | 235.4             | 2.5 a                            | 2.3 a | 2.8               |
| 'Flying Dragon'                          | 54.6              | 47.6              | 58.0              | 260.9 b | 307.4 c       | 225.5             | 2.6 a                            | 2.2 a | 2.9               |
| 'Rangpur' lime                           | 57.9              | 51.8              | 58.6              | 323.4 a | 339.0 b       | 240.7             | 2.1 b                            | 1.9 b | 2.8               |
| 'Florida' rough lemon                    | 60.7              | 48.8              | 59.0              | 312.3 a | 382.3 a       | 245.5             | 2.1 b                            | 1.7 b | 2.7               |
| 'Sunki' tangerine                        | 62.1              | 53.0              | 58.0              | 271.2 b | 293.7 с       | 230.7             | 2.5 a                            | 2.3 a | 2.9               |
| F–value                                  | 1.6 <sup>ns</sup> | 1.6 <sup>ns</sup> | 1.1 <sup>ns</sup> | 3.1 *   | 4.4 *         | 1.2 <sup>ns</sup> | 3.0 *                            | 5.0 * | 1.4 <sup>ns</sup> |

Table 5. Industrial properties of juices of 'Valencia' sweet orange trees grafted on 20 rootstocks.

Means within columns followed by the same letter do not differ statistically by one-way ANOVA with the rootstock as variability factor (Scott–Knott test, p < 0.05). \* p < 0.01. ns: not significant. <sup>a</sup>: No. of boxes of 40.8 kg per ton of frozen concentrated orange juice; <sup>b</sup>: kg of SS per box of 40.8 kg.

In the 2019 harvest season, there were no significant differences among rootstocks in terms of juice yield, industrial yield, technological index and TA. Low SS contents were observed for the fruits of trees grown on 'US–801' citrandarin (11.4 °Brix), 'Murcott' tangor  $\times$  Trifoliata–9' (11.7 °Brix), 'Rangpur' lime (11.7 °Brix), 'Florida' rough lemon (11.2 °Brix) and on all of the citrumelo rootstocks (11.1 to 11.9 °Brix), but not for those on 'Swingle' rootstocks. Moreover, the fruits of trees grown on 'US–852' citrandarin and 'Trifoliata' rootstocks showed the highest mean of MI (24.4 and 22.9, respectively) (Tables 4 and 5).

#### 3.4. Principal Component Analysis

The Principal Component Analysis (PCA) was successfully applied to identify groupings in relation to the variables assessed, where >70% of the variance was explained by the first and second principal components (53.4% and 22.8%, respectively). Moreover, agglomerative hierarchical clustering grouped the rootstock genotypes based on tree development, production and fruit quality (Figure 1). Industrial yield and technological index, which were negatively associated, provided the greatest contribution to PC1; they enabled the identification of three distinct rootstock groups. The first group comprised the 'US–812', IPEACS–264, IPEACS–239 and 'US–852' citrandarins, 'Trifoliata' and 'Flying Dragon' trifoliate, with fruits of superior quality, technological index and industrial yield.



**Figure 1.** Principal components analysis (PCA) of production (PROD), canopy volume (CV), production efficiency (PE), technological index (TI), soluble solids (SS), maturation index (SS/TA), industrial yield (IY) and juice yield (JY) of 'Valencia' sweet orange grafted onto 20 rootstocks. (**A**). Treatment dispersion according to the scores of the principal components. (**B**). Variable arrangement according to the scores of the principal components. US852Citrand: 'US-852' citrandarin; US801Citrand: 'US-801' citrandarin; US812Citrand: 'US-812' citrandarin; IPEACS239Citrand: IPEACS-239 citrandarin; IPEACS256Citrand: IPEACS-256 citrandarin; IPEACS264Citrand: IPEACS-264 citrandarin F803Citrum: F.80–3 citrumelo; F805Citrum: F.80–5 citrumelo; F806Citrum: F.80–6 citrumelo; F807Citrum: F.80–7 citrumelo; F80–8Citrum: F.80–8 citrumelo; W2Citrum: 'W-2' citrumelo; SwiCitrum: 'Swingle' citrumelo; US802: 'US-802' pummelo hybrid; MurTanTri9: 'Murcott' tangor × 'Trifoliata'-9; Trif: 'Trifoliata'; FlyDr: 'Flying Dragon'; RangLim: 'Rangpur' lime; FloRouLem: 'Florida' rough lemon; SunkTang: 'Sunki' tangerine.

Conversely, the group comprising the 'Rangpur' lime, 'Florida' rough lemon and F.80–7 citrumelo had fruits of lower quality. The other rootstocks evaluated, comprising the third and largest group, had an intermediate fruit-quality. Moreover, yield and canopy

volume were positively associated, revealing that the vigorous rootstocks also had higher yield, but with lower production efficiency.

# 4. Discussion

Smaller tree size and canopy volume are key characteristics attributed to 'Trifoliata' hybrid rootstocks [26,29,31]. However, the growth development characteristics were highly variable among our assessed hybrids, especially when comparing genotypes from the same progenitor rootstocks (for instance, citrumelos and citrandarins). This indicates that these variables may be under the genetic control of several heterozygous *loci* [40], or that these genotypes carry commercially unattractive characteristics of the parents, as we observed for the trees grown on 'Sunki' and 'Cleopatra' tangerines, which normally have canopies with high vegetative vigor [9,28].

In our study, the trees grown on at least 10 of the 'Trifoliata' hybrid rootstocks were of low vigor, in particular IPEACS–239 and IPEACS–256 citrandarins, US–802 pummelo hybrid and F.80–3 and F–80.5 citrumelos (Table 2). On average, the canopy volume of these rootstocks at the last assessment was  $\approx$ 100% and 60% lower than the commercial rootstocks 'Rangpur' lime and 'Swingle' citrumelo, respectively.

Currently, breeding programs aim to obtain less vigorous rootstocks, that is, those with dwarfing potential [2,30,41]. Low-vigor orchards tend to be more profitable because they enable high-density planting, favor manual and mechanical harvesting operations, allow more efficient control of pests and diseases and facilitate other agricultural treatments, such as spraying, fertilization and pruning [22,42]. It is important to note that in high-density planting, the initial investment tends to be higher because more trees are required. Therefore, it is crucial that scion/rootstock combinations that are suitable for high-density planting systems ensure orchard profitability, and cover the costs of establishment in the first bearing seasons.

Further, highly vigorous trees require more manual labor to harvest, because it is difficult to collect fruit from higher in the canopy, often requiring the use of ladders and harvesting hooks; this reduces the operating yield of orange pickers [22,43]. Mechanical harvesting is increasingly being proposed as a solution, and has been implemented in several countries, with new models of orange harvesters being developed and made available on the market. It is likely that this practice, which aims to optimize harvesting and minimize production costs by reducing labor, represents the future of the citrus industry [43–45]. Establishing new orchards with orange trees grafted on dwarfing rootstocks, as observed in our trial, can favor mechanical harvesting, because the reduced canopy expansion between rows allows for more efficient traffic of large machines and other agricultural machinery, facilitates harvesting and avoids damage to the trees and to the fruits.

Canopy overlap between rows hinders management practices and harvesting operations such as the transit of pickers, placement of harvest boxes for the temporary storage of fruit and the entry of agricultural machinery [22]. High-density planting therefore requires frequent pruning to avoid this, and scion/rootstock combinations with shorter and narrower canopies may reduce the need for pruning in the early years of the orchard. For instance, we observed that the trees grafted onto F.80–3 and F.80–5 citrumelos, IPEACS– 256 and IPEACS–239 citrandarins and 'US–802' pummelo hybrid had reduced vegetative vigor, slowing down canopy overlap and the formation of a continuous hedgerow. Thus, these genotypes are potential alternative rootstocks for 'Valencia' orange trees grown in high-density planting, because they produced dwarf trees with narrow and small canopies.

Earliness is a desired characteristic in new orange plantings. In our trial, 'Valencia' orange trees grown on the 'US–812' citrandarin, 'W–2' citrumelo and 'Rangpur' lime produced earlier than the other rootstocks evaluated, whereas those on the other 'Trifoliata' hybrid rootstocks produced later and more regularly, and were more likely to show increased production in later harvest seasons. 'Valencia' orange trees grown on 'W–2' citrumelo rootstock have earlier and higher production than those grown on 'Rangpur' lime, 'Hamlin' [39] and 'Valencia' trees on 'US–812' citrandarin rootstock have good productive performance, with high production in the early bearing seasons [10,30,33].

Orange varieties grafted on the 'Rangpur' lime tend to have higher and early production, characteristics that make it one of the most used rootstocks in Brazil [8,9]. The use of early producing scion/rootstock combinations, i.e., those bearing high yields in the first crops, as we observed for 'Valencia' orange trees on 'US–812' citrandarin and 'W–2' citrumelo rootstocks, is a good strategy for ensuring financial return and covering the initial costs when establishing a new orchard. However, it is important to select rootstocks that promote regular production and ensure the longevity of the orchard.

Orange production is directly related to tree growth development [7,10,30,31]. In our trial, trees on F.80–3 and F.80–5 citrumelos, IPEACS–256 and IPEACS–239 citrandarins and 'US–802' pummelo hybrid had low yield, mainly due to their dwarfing potential. The selection of scion/rootstock combinations requires clearly defined cultivation strategies for each growing region, as a mean to select genotypes more likely to generate profits over the life cycle of the orchard. The most productive rootstocks are not always the best alternatives; for instance, although the 'Rangpur' lime is a high yielding rootstock, it is also highly vigorous, which is commercially undesirable [2,4,41]. Under our study conditions, the 'US–801' and 'US–812' citrandarin rootstocks were promise, with good productive performance and high and regular production over the study; further, they produced less vigorous trees than the rootstocks grown on a commercial scale.

Production efficiency is currently a determining factor when choosing scion/rootstock combinations, given the strong tendency toward high-density planting, which prioritizes increased yield [22,41]. Rootstocks with dwarfism potential and supporting high production efficiency are good options under high-density planting. [20,24]. In this trial, the production efficiency of 'Valencia' orange trees on F.80–3 and F.80–5 citrumelos, 'US–802' pummelo hybrid and on IPEACS–239 and IPEACS–256 citrandarins was on average  $\approx$ 55% higher than on 'Rangpur' lime, one of the most used rootstocks in Brazil.

Among the rootstocks with high production efficiency, those with lower vigor are preferred, as they increase yield by allowing high-density planting, facilitate efficient orchard management and minimize production costs [10,22,46,47]. Therefore, under high-density planting, the increase in yield should compensate for the initial high expenses of establishing the orchard, and should ensure the long-term economic return.

Orchards established with vigorous rootstocks have higher production losses when diseased trees are removed, because the production per tree is usually higher than that of less vigorous rootstocks, i.e., those with dwarfing potential [2,10,48]. For instance, considering that 20% of the 'Valencia' trees in an orchard are removed, trees grown on 'Florida' rough lemon rootstock would have higher productivity losses than those grown on low vigor rootstocks with higher production efficiency (namely, F.80–3 and F.80–5 citrumelo, IPEACS–239 citrandarin and 'US–802' pummelo hybrid).

Therefore, high-density plantings of rootstocks with high production efficiency and low vigor, such as the F.80–3 and F.80–5 citrumelo and the IPEACS–239 citrandarin, provide an important means to increase orchard yield, provided that the higher density does not reduce production by necessitating severe pruning and removal of diseased trees. Further, high-density plantings minimize the costs of phytosanitary treatments, manual and mechanical harvesting operations and other agricultural practices. Moreover, these benefits are fundamental to ensuring the maintenance of small- and medium-sized citrus farms, provided that the management practices involved are not overly technological. These farms require effective and low-cost strategies to keep operating.

In terms of industrial yield, trees on IPEACS–239 citrandarin rootstock would require up to  $\approx 20\%$  fewer 40.8 kg boxes to obtain a ton of concentrated juice than those grown on the 'Rangpur' lime, and up to  $\approx 30\%$  fewer boxes than those grown on the 'Florida' rough lemon rootstock. These two rootstocks are inferior to various 'Trifoliata' hybrids in terms of industrial yield, technological index and SS content [9,49–51]. In this trial, most of the 'Trifoliata' hybrid rootstocks induced better chemical and industrial properties of 'Valencia' orange juice than 'Rangpur' lime.

The choice of rootstocks with good fruit quality, industrial yield and technological index, as for the 'Valencia' trees grown on IPEACS–239, IPEACS–256 and 'US–812' citrandarin rootstocks, is essential for increasing profits throughout the citrus industry. The use of rootstocks providing poor fruit quality—such as the 'Rangpur' lime and the 'Florida' rough lemon—is still significant, especially in countries where the growers are paid by fruit weight rather than the quantity of SS.

In our trial, the rootstocks already used on a commercial scale, such as the 'Rangpur' lime, 'Florida' rough lemon, 'Sunki' tangerine and 'Flying Dragon' trifoliate, produced inferior results in terms of tree growth, productive performance and industrial properties of orange juices than some of the 'Trifoliata' hybrids. Thus, we were able to identify promising genotypes for the diversification of citrus orchards, such as IPEACS–239, IPEACS–256 and 'US–812' citrandarins, 'US–802' pummelo hybrid and the F.80–3 and F.80–5 citrumelos. These rootstocks produce high-quality fruit, and allow harvesting operations and strategies that minimize costs, including phytosanitary treatments, in addition to increasing yields via high-density planting.

Based on our findings, a Quick Reference Chart (Table S1) was created for the practical and objective identification of the best rootstock alternatives for 'Valencia' orange trees. This chart will assist with decision-making under various cultivation scenarios, helping citrus farmers to achieve greater production efficiency and profits. For example, the IPEACS–239 and IPEACS–256 citrandarins and the 'US–802' pummelo hybrid are excellent alternatives for high-density planting of 'Valencia' orange trees, with low vigor, good production and high production efficiency and industrial properties of orange juice; they are superior to the other rootstocks that are also appropriate for this scenario. Conversely, under low density planting, 'Rangpur' lime can be replaced by the 'US–812' citrandarin as the rootstock, or even with 'US–801' or 'Murcott' tangor × 'Trifoliata'–9, which induces trees of intermediate vigor, optimal production and optimum industrial properties of orange juices.

# 5. Conclusions

'Valencia' orange trees grown on IPEACS–239 and IPEACS–256 citrandarins, and on 'US–802' pummelo hybrid have dwarfing potential, high production efficiency and high industrial properties of orange juice and are therefore alternatives for high-density plantings. The F.80–3 and F.80–5 citrumelos also have good dwarfing potential and high production efficiency, but lower industrial properties of juice compared to the other 'Trifoliata' hybrid rootstocks.

Trees grown on 'US–812' citrandarin rootstock has low vigor, good productive performance, accumulated production and production efficiency similar to 'Rangpur' lime, and high industrial properties of juices. Although the 'Rangpur' lime and the 'Florida' rough lemon allow high yield, the trees are very vigorous, with low-quality fruits. The Quick Reference Chart created enables practical and objective identification of the best rootstock alternatives for 'Valencia' orange trees in terms of tree growth development and industrial properties of juices.

**Supplementary Materials:** The following are available online at https://www.mdpi.com/article/ 10.3390/horticulturae7060141/s1, Table S1: Quick Reference Chart describing the influence of 20 rootstock genotypes on canopy volume, production, production efficiency, technological index, and industrial yield of the 'Valencia' sweet orange.

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# References

- 1. Ramin, A.A.; Alirezanezhad, A. Effects of citrus rootstocks on fruit yield and quality of Ruby Red and Marsh grapefruit. *Fruits* **2005**, *60*, 311–317. [CrossRef]
- 2. Castle, W.S. A career perspective on citrus rootstocks, their development, and commercialization. *HortScience* **2010**, *45*, 11–15. [CrossRef]
- 3. Legua, P.; Bellver, R.; Forner, J.; Forner-Girner, M.A. Plant growth, yield and fruit quality of 'Lane Late' navel orange on four citrus rootstocks. *Span. J. Agric. Res.* 2011, *9*, 271–279. [CrossRef]
- 4. Martínez-Cuenca, M.R.; Primo-Capella, A.; Forner-Giner, M.A. Influence of Rootstock on Citrus Tree Growth: Effects on Photosynthesis and Carbohydrate Distribution, Plant Size, Yield, Fruit Quality, and Dwarfing Genotypes. In *Plant Growth*; Rigobelo, E.C., Ed.; IntechOpen: Rijeka, Croatia, 2016; Chapter 8. [CrossRef]
- 5. França, N.O.; Amorim, M.S.; Girardi, E.A.; Passos, O.S.; Soares Filho, W.S. Performance of 'Tuxpan Valencia' sweet orange grafted onto 14 rootstocks in northern Bahia, Brazil. *Rev. Bras. Frutic.* **2016**, *38*, e684. [CrossRef]
- Kirinus, M.B.M.; Oliveira, R.P.; Silva, P.S.; Barreto, C.F.; Malgarim, M.B.; dos Santos Soares Filho, W. Agronomic performance of 'Valencia' orange combined with 13 rootstocks in the State of Rio Grande do Sul, Brazil. *Pesqui. Agropecuária Bras.* 2019, 54, e01349. [CrossRef]
- Carvalho, L.M.; Carvalho, H.W.L.; Barros, I.; Martins, C.R.; dos Santos Soares Filho, W.; Girardi, E.A.; Passos, O.S. New scion-rootstock combinations for diversification of sweet orange orchards in tropical hard setting soils. *Sci. Hortic.* 2019, 243, 169–176. [CrossRef]
- 8. Pompeu Júnior, J.; Blumer, S. Performance de citrumelos F–80 no Estado de São Paulo. Laranja 2005, 26, 77–85.
- 9. Domingues, A.R.; Neves, C.S.V.J.; Yada, I.F.U.; Leite-Junior, R.P.; Tazima, Z.H. Performance of 'Cadenera' orange trees grafted on five rootstocks. *Rev. Bras. Frutic.* **2018**, 40, e764. [CrossRef]
- Fadel, A.L.; Stuchi, E.S.; Couto, H.T.Z.; Ramos, Y.C.; Mourão Filho, F.A.A. Trifoliate hybrids as alternative rootstocks for 'Valencia' sweet orange under rainfed conditions. *Sci. Hortic.* 2018, 235, 397–406. [CrossRef]
- 11. Rodrigues, M.J.S.; Ledo, C.A.S.; Girardi, E.A.; Almeida, L.A.H.; Soares Filho, W.S. Caracterização de frutos e propagação de porta-enxertos híbridos de citros em ambiente protegido. *Rev. Bras. Frutic.* **2015**, *37*, 457–470. [CrossRef]
- Rodrigues, M.J.S.; Neto, R.C.A.; Neto, S.E.A.; dos Santos Soares Filho, W.; Girardi, E.A.; Lessa, L.S.; Almeida, U.O. Performance of 'Valencia' sweet orange grafted onto rootstocks in the State of Acre, Brazil. *Pesqui. Agropecuária Bras.* 2019, 54, e1349. [CrossRef]
- 13. Hutchison, D.J. Swingle citrumelo—A promising rootstock hybrid. Proc. Fla. State Hortic. Soc. 1974, 87, 89–91.
- Castle, W.S.; Wutscher, H.K.; Youtsey, C. Citrumelos as rootstocks for Florida citrus. *Proc. Fla. State Hortic. Soc.* 1988, 101, 28–33.
  Bowman, K.D.; Graham, J.H.; Adair, R.C., Jr. Young tree growth in a flatwoods rootstock trial with Diaprepes weevil and Phytophthora diseases. *Proc. Fla. State Hortic. Soc.* 2003, 116, 249–251.
- Girardi, E.A.; Cerqueira, T.S.; Cantuarias-Avilés, T.E.; Silva, S.R.; Stuchi, E.S. Sunki mandarin and Swingle citrumelo as rootstocks for rain-fed cultivation of late-season sweet orange selections in northern São Paulo state, Brazil. *Bragantia* 2017, 76, 501–511. [CrossRef]
- 17. Barry, G.H.; Castle, W.S.; Davies, F.S. Soluble solids accumulation in 'Valencia' sweet orange as related to rootstock selection and fruit size. *J. Am. Soc. Hortic. Sci.* 2004, 129, 594–598. [CrossRef]
- 18. Spreen, T.H. The word citrus industry. In *Soils, Plant Growth and Crop Production*, 1st ed.; Verheye, W.H., Ed.; UNESCO/Eolss Publishers: Singapore, 2010; pp. 249–269.
- 19. Domingues, A.R.; Marcolini, C.D.M.; Gonçalves, C.H.S.; Gonçalves, L.S.A.; Roberto, S.R.; Carlos, E.F. Fruit ripening development of 'Valencia' orange trees grafted on different 'Trifoliata' hybrid rootstocks. *Horticulturae* **2021**, *7*, 3. [CrossRef]
- 20. Cantuarias-Avilés, T.; Mourão Filho, F.A.A.; Stuchi, E.S.; Silva, S.R.; Espinoza-Nuñez, E.; Neto, H.B. Rootstocks for high fruit yield and quality of 'Tahiti' lime under rain-fed conditions. *Sci. Hortic.* **2012**, *142*, 105–111. [CrossRef]
- Forner-Giner, M.A.; Rodriguez-Gamir, J.; Martinez-Alcantara, B.; Quiñones, A.; Iglesias, D.J.; Primo-Millo, E.; Forner, J. Performance of 'Navel' orange trees grafted onto two new dwarfing rootstocks (Forner-Alcaide 517 and Forner-Alcaide 418). *Sci. Hortic.* 2014, 179, 376–387. [CrossRef]
- Stuchi, E.S.; Girardi, E.A.; Sempionato, O.R.; Reiff, E.T.; Silva, S.R.; Parolin, L.G. Trifoliata 'Flying Dragon': Porta–Enxerto para Plantios Adensados e Irrigados de Laranjeiras Doces de alta Produtividade e Sustentabilidade; Comunicado Técnico, 152; Embrapa Mandioca e Fruticultura Tropical: Cruz das almas, Brazil, 2012; 6p.
- 23. Sulzbach, M.; Oliveira, R.P.; Girardi, E.A.; Schwarz, S.F.; Bertolini, E.; Schneider, L.A.; Gonzatto, M.P. *Huanglongbing (HLB) dos Citros e Estratégias de Manejo Visando Prevenção e Controle*; Comunicado Técnico, 450; Embrapa Clima Temperado: Pelotas, Brazil, 2017; 26p.
- 24. Donadio, L.C.; Lederman, I.E.; Roberto, S.R.; Stuchi, E.S. Dwarfing-canopy and rootstock cultivars for fruit trees. *Rev. Bras. Frutic.* **2019**, *41*, e997. [CrossRef]

- 25. Vashisth, T.; Chun, C.; Hampton, M.O. Florida citrus nursery trends and strategies to enhance production of field-transplant ready citrus plants. *Horticulturae* **2020**, *6*, 8. [CrossRef]
- 26. Castle, W.S.; Tucker, D.P.H.; Krezdorn, A.H.; Youtsey, C.O. *Rootstocks for Florida Citrus*, 2nd ed.; University of Florida: Gainesville, FL, USA, 1993.
- 27. Bowman, K.D.; Rouse, R.E. US-812 Citrus rootstock. HortScience 2006, 41, 832-836. [CrossRef]
- Schinor, E.H.; Cristofani-Yaly, M.; Bastianel, M.; Machado, M.A. Sunki Mandarin vs *Poncirus trifoliata* hybrids as rootstocks for Pera sweet orange. J. Agric. Sci. 2013, 5, 190–200. [CrossRef]
- 29. Andersen, P.C.; Brodbeck, B.V. Yield, tree size, and fruit quality of mature 'Owari' and 'Brown Select' satsuma on Poncirus trifoliata 'Rubidoux' and 'Flying Dragon' rootstocks in north Florida. *HortScience* **2015**, *50*, 1650–1653. [CrossRef]
- 30. Bowman, K.D.; McCollum, G. Five new citrus rootstocks with improved tolerance to huanglongbing. *HortScience* 2015, 50, 1731–1734. [CrossRef]
- 31. Bowman, K.D.; McCollum, G.; Albrecht, U. Performance of 'Valencia' orange [*Citrus sinensis* (L.) Osbeck] on 17 rootstocks in a trial severely affected by huanglongbing. *Sci. Hortic.* **2016**, 201, 355–361. [CrossRef]
- Alvares, C.A.; Stape, J.L.; Sentelhas, P.S.; de Moraes Gonçalves, J.L.; Sparovek, G. Köppen's climate classification map for Brazil. *Meteorol. Z.* 2013, 22, 711–728. [CrossRef]
- 33. Nitsche, P.R.; Caramori, P.H.; da Silva Ricce, W.; Pinto, L.F.D. Atlas Climático do Estado do Paraná; IAPAR: Londrina, Brazil, 2019.
- Mattos Junior, D.; Bataglia, O.C.; Quaggio, J.A. Nutrição dos Citros; Mattos Junior, D., de Negri, J.D., Pio, R.M., Pompeu Junior, J.C., Eds.; Fundag; Instituto Agronômico: Campinas, Brazil, 2005; pp. 197–219.
- 35. Carvalho, J.E.B.; de Neves, C.S.V.J.; Menegucci, J.L.P.; Silva, J.A.S. *Práticas Culturais*; Mattos Junior, D., de Negri, J.D., Pio, R.M., Pompeu Junior, J.C., Eds.; Fundag; Instituto Agronômico: Campinas, Brazil, 2005; pp. 449–482.
- 36. Mendel, K. Rootstock-scion relationships in Shamouti trees on light soil. Ktavim 1956, 6, 35-60.
- 37. Association of Official Analytical Chemists. *Official methods of analysis*, 15th ed.; Association of Official Analytical Chemists: Arlington, VA, USA, 1990.
- 38. Ferreira, D.F. Sisvar: A computer statistical analysis system. Ciência Agrotecnologia (UFLA) Lavras 2011, 35, 1039–1042. [CrossRef]
- Pompeu, J., Jr.; Blumer, S. Citrumelos como porta-enxertos para laranja 'Valência'. Pesq. Agropec. Bras. 2011, 46, 105–107. [CrossRef]
- 40. Bordignon, R.; Medina Filho, H.P.; Siqueira, W.J.; Pio, R.M. Características da laranjeira 'Valencia' sobre clones e híbridos de porta-enxertos tolerantes à tristeza. *Bragantia* 2003, *62*, 381–395. [CrossRef]
- 41. Ladaniya, M.S.; Marathe, R.A.; Das, A.K.; Rao, C.N.; Huchche, A.D.; Shirgure, P.S.; Murkute, A.A. High density planting studies in acid lime (Citrus aurantifolia Swingle). *Sci. Hortic.* **2019**, *261*, 108935. [CrossRef]
- 42. Carlos, E.F.; Stuchi, E.S.; Donadio, L.C. *Porta-Enxertos Para a Citricultura Paulista*; Boletim Citrícola n.1; Funep: Jaboticabal, Brazil, 1997; 47p.
- 43. Ferreira, M.D.; Sanchez, A.C.; Braumbeck, O.A.; Santos, E.A. Harvesting fruits using a mobile platform: A case study applied to citrus. *Eng. Agric.* 2018, *38*, 293–299. [CrossRef]
- 44. Sanders, K.F. Orange Harvesting Systems Review. Biol. Eng. 2005, 90, 115–125. [CrossRef]
- Roka, F.M.; Hyman, B.R. Mechanical harvesting of sweet oranges for juice processing. *Acta Hortic.* 2012, *965*, 241–244. [CrossRef]
  Teófilo Sobrinho, J.; Pompeu Junior, J.; Figueiredo, J.O. Adensamento de plantio da laranjeira 'Valencia' sobre Trifoliata. *Laranja*
- 2012, 33, 49–58.
- 47. Mademba-Sy, F.; Lemerre-Desprez, Z.; Lebegin, S. Use of Flying Dragon trifoliate orange as dwarfing rootstock for citrus under tropical climatic conditions. *HortScience* **2012**, *47*, 11–17. [CrossRef]
- 48. Carvalho, S.A.; Girardi, E.A.; Mourão Filho, F.A.A.; Ferrarezzi, R.S.; Coletta Filho, H.D. Advances in citrus propagation in Brazil. *Rev. Bras. Frutic.* **2019**, *41*, e422. [CrossRef]
- 49. Auler, P.A.M.; Fiori-Tutida, A.C.G.; Tazima, Z.H. Comportamento da laranjeira 'Valencia' sobre seis porta–enxertos no Noroeste do Paraná. *Rev. Bras. Frutic.* 2008, *30*, 229–234. [CrossRef]
- 50. Castle, W.S.; Baldwin, J.C.; Murano, R.P. Performance of 'Valencia' sweet orange trees on 12 rootstocks at two locations and an economic interpretation as a basis for rootstock selection. *HortScience* **2010**, *45*, 523–533. [CrossRef]
- 51. Amorim, M.S.; Girardi, E.A.; França, N.O.; Gesteira, A.S.; dos Santos Soares Filho, W.; Passos, O.S. Initial performance of alternative citrus scion and rootstock combinations on the northern coast of the state of Bahia, Brazil. *Rev. Bras. Frutic.* **2018**, 40, e480. [CrossRef]