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Tree Performance, Yield, and Fruit Quality of 'Valencia' Sweet Orange (*Citrus sinensis* L. Osbeck) Selections on New *Poncirus trifoliata* Rootstocks

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Abstract: This study reports the performance of seven Valencia (*Citrus sinensis*) selections grafted to four Trifoliata (*Poncirus trifoliata)* rootstocks in the Riverina region of south-east Australia. Six of the Valencia selections (numbered as Valencia 1 to 6) were from orchards in the Riverina region, and the seventh was a standard commercial variety 'Keenan' (control). Three of the four Trifoliata rootstock selections ('Zao Yang', 'Tanghe', and 'Donghai') were imported from the People's Republic of China, and the fourth was the 'Tri22' (control) selection used commercially in Australia. 'Valencia 5' produced the highest cumulative (162 kg/tree) yield over the five harvest seasons and increased levels of Total Soluble Solids (TSS, 1.7 °Brix) compared with the current industry standard, 'Keenan'. Trees grafted to 'Zao Yang' produced higher yields than any of the other Trifoliata selections, again outperforming the current industry standard, 'Tri22'. The new combination of 'Valencia 5' scion on 'Zao Yang' rootstock represents an opportunity to significantly improve orchard productivity, particularly for juice production. 'Valencia 6' on Zao Yang' had the highest percentage (58%) of fruit >75 mm in diameter compared with other scion/rootstock combinations and creates an additional opportunity for fresh market production. Significant differences in tree size, growth rates, and productivity were identified. Results from this evaluation have resulted in the commercialisation of 'Valencia 5' and 'Valencia 6' scions and 'Zao Yang' rootstock as three new citrus varieties for sweet orange production in Australia.

Keywords: total soluble solids; juice%; yield efficiency; alternate bearing index; trunk circumference

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1. Introduction

The citrus industry is one of Australia's largest fresh produce exporters, with export volumes increasing 84% from 158,000 tonnes in 2014 to over 222,000 tonnes in 2022, planted over 29,000 ha [1]. Thirty percent of Australian citrus is grown in the Riverina region of Australia, with Valencia orange [*Citrus sinensis* (L.) Osb.] being the dominant variety. Total production in the 2022 season for Valencia orange in the Riverina was 140,000 tonnes over 4233 hectares [1]. Riverina plantings are predominantly grown on 'Tri22' rootstock, the Australian selection of *Poncirus trifoliata* [1].

The citrus industry has thrived in the Riverina since the 1950s. A significant proportion of the orchards were reaching the end of their productive life; therefore, it was suggested that productive local selections may be identified in the old blocks for future replanting. This would ensure the continued supply of high-quality Valencia fruit in sufficient volumes to meet the processing needs of the local citrus industry in the Riverina.

Six Valencia orchard blocks, predominantly established in the 1950s, were selected from long-term production records by National Foods Limited, Leeton. A budwood source tree was selected in each block. Budwood of each scion variety was transferred to Dareton

Primary Industries Institute in the Sunraysia citrus growing region, and mother trees were established to supply budwood material for propagation of trial trees.

Chinese rootstocks were imported as seed from the Citrus Research Institute South-West University, Chongqing, People's Republic of China, in 1992 as part of an Australian Centre for International Agricultural Research (ACIAR) project [2]. These rootstocks were evaluated in field trials at the Primary Industries Institute, Dareton, New South Wales, for 10 years in deep sandy loam soils, typical of those in the Sunraysia region [3], and the six high-performing rootstocks were released for commercial use in Australia based on yield, quality, and graft union compatibility [4]. Three of these six new rootstocks were pure Trifoliata selections [2] and were identified for field evaluation in the Riverina to test their performance in sandy clay soil conditions and in combination with the recently selected Valencia clones.

Rootstock variety is reported to significantly influence the entire grafted tree, particularly vigour, productivity, fruit quality, and resistance to abiotic and biotic stress [5]. In a previous study, it was demonstrated that there was a six-fold yield difference between the best- and worst-performing rootstocks used for Valencia scions [6]. Internal fruit quality, measured by sugar content, acidity, juiciness, and taste, is important for both fresh consumption and processing oranges [7]. However, sweetness and juice percent are markedly important for the juicing industry. Sweetness is often governed, in part, by sugar concentration [8]. Therefore, the determination and quantification of sugars is of great importance for the processing industry [9,10].

Fruitfulness or productiveness of Valencia selections, expressed as production of fruit or sugar total soluble solids (TSS) per unit area of tree canopy, demonstrates the suitability of selections to fresh fruit marketing and juice processing [11]. Replicated field evaluation over a long timeframe is the ideal, but there is pressure to provide short-term comparative data [12]. Relative rootstock performance with the Hamlin processing orange variety in Florida demonstrated that reliable conclusions could be made based on results from the first four cropping years [13].

This work reports the response of seven Valencia selections established on four different *Poncirus trifoliata* rootstocks in a field trial in the Riverina, NSW, to assess tree growth, yield, fruit quality, and fruit size. Data from the first five cropping years are presented. The aim of this work was to identify a superior Valencia scions and Trifoliata rootstock combination for the Australian juicing industry.

2. Materials and Methods

2.1. Plant Material

Budwood of six Valencia scion selections was collected from six citrus orchards in the Riverina region of NSW in January 2005. The six Valencia selections were labelled as 'Valencia 1', 'Valencia 2', 'Valencia 3', 'Valencia 4', 'Valencia 5', and 'Valencia 6'. A seventh Valencia selection, 'Keenan', was included as a control, and is the long-term and current industry standard in Australia.

Buds from the selected source trees were grafted to *Troyer citrange* rootstocks to establish mother trees at Dareton Primary Industries Institute. The health status of the mother trees was established by sending material for biological and serological testing at the Elizabeth Macarthur Agricultural Institute, Menangle, NSW. Biological indexing was undertaken for citrus exocortis (CEV) viroid on 'Etrog' citron indicator plants, citrus tristeza virus (CTV) on West Indian lime and Eureka lemon, citrus psorosis virus (CPsV) and CPsV-like viruses on Symons sweet orange and Emperor mandarin, and citrus tatter leaf virus (CTLV) on Rusk citrange [14]. CTV was detected using a direct tissue blot immunoassay [15].

All mother trees were determined to be free of CEV, CPsV, CPsV-like, and CTL pathogens. CTV was detected in five of the seven Valencia selections but was determined to be mild isolates that have no impact on scion performance when used with CTV-resistant rootstocks [16]. 'Valencia 5' and 'Keenan' (control) mother trees were free of

CTV. In summary, the budwood source trees were uniformly free of pathogens that can cause disease in Valencia sweet orange when grown on Trifoliata rootstock [16]. During the entire period of study, all trees were regularly inspected and observed to be equally healthy throughout the nursery propagation phase, during the field trial period, and to the present.

Rootstock fruit was collected, and the seed extracted, from source trees of the Chinese rootstocks ‘Donghai’, ‘Tanghe’, and ‘Zao Yang’ established at Dareton. Seed of the Australian standard *Poncirus trifoliata* rootstock ‘Tri22’ (control) was sourced from the Auscitrus (national accredited budwood and seed scheme). Valencia selections were grafted to the 4 rootstocks and grown in a protected shade house over a 2-year period before planting.

2.2. Field Layout and Experimental Design

The trial site was located on a commercial property at Stanbridge (Latitude. 34.5175° S, Longitude. 146.2356° E) near Leeton in the Riverina region of NSW, approximately 550 km west of Sydney, Australia. The trial site was cleared of 25-year-old citrus orchard, cultivated, and left fallow for 12 months prior to planting. Five tonnes of cow manure and three tonnes of gypsum were applied per hectare, evenly spread, and worked into the soil prior to forming 60 cm high mounds.

An EM38 (electromagnetic soil mapping) survey was conducted on the trial site to determine soil electrical conductivity (EC) at designated depths. EC is strongly influenced by soil texture, clay, and salt content. The EM38 survey assisted the site selection and extraction of five 120 cm cores over the 0.4 ha site to assist soil characteristics and support the experimental design. The soil textures were predominantly sandy loam in the 0–10 cm layer, sandy clay from 10–40 cm, and light clay from 40–120 cm. The 60-cm-high flat topped planting mounds were hand cored to assess soil characteristics prior to planting. Particle size analysis gave a clay content (<2 μm) of 17%, silt (>2–<20 μm) of 8%, fine sand (20–200 μm), and coarse sand (200–2000 μm) of 47%. Soil pH was consistent at 7.5 ($CaCl_2$) at 0–20 cm and 20–60 cm depths. Soil electrical conductivity ranged from 0.10–0.35 dS/m within the planting mound.

Trees were planted in October 2010 at a spacing of 1.8 m within the rows and 5 m between rows in an east–west orientation, creating a density of 1111 trees/ha. The high-density planting was established with the future aim of mechanised fruit harvest of Valencia oranges. The experiment was a randomised complete block design with 2 treatment factors (7 Valencia selections × 4 rootstocks) replicated 3 times. There were 5 trees per plot, giving a trial size of 420 trees. The site was buffered by two guard rows on each side and an individual tree at both ends of each of the seven trial rows.

The typical dry semi-arid climate of south-east Australia requires irrigation for crop production. The trees were initially irrigated via a single drip line with a second line added in year 3 from planting. Irrigation scheduling was based on gypsum block sensors established at 25 cm and 50 cm soil depths and mid-way between drip emitters within the planting mound. The sensors were linked to a GDot visual display with an MEA switch box (www.mea.com.au, accessed on 15 March 2024). Fertiliser was applied via the drip system, and standard commercial cultural practices were followed during the experiment. Trees were lightly side pruned during the experiment to allow easy access for farm machinery and spray operations.

2.3. Climatic Conditions

The average daily maximum temperature at Stanbridge ranges from 30–32 °C during December–February and 15–17 °C during June–August, with an average annual rainfall of 420 mm. Annual accumulated heat units average 1800 (calculated as [Maximum temperature + Minimum temperature]/2 − 13 °C (maximum threshold of 35 °C) [17]. Detailed climatic data throughout the experimental trial are given in (Table 1).

Table 1. The total number of days of minimum and maximum temperature extremes for 5 growing seasons (December–January) across a range of temperatures.

Temperature Range (°C)	2015	2016	2017	2018	2019
		Minimum temperatures (May–September)			
<0	23	13	44	26	32
0 to −1	13	11	15	11	18
−1.1 to −2	7	2	6	6	10
−2.1 to −3	3	0	16	4	4
−3.1 to −4	0	0	4	5	0
−4.1 to −5	0	0	3	0	0
		Maximum temperatures (October–April)			
>35	40	42	42	44	62
35.1 to 40	31	37	35	31	36
40.1 to 45	9	5	7	13	22
45.1 to 50	0	0	0	0	4

2.4. Data Collection and Analyses

2.4.1. Vegetative Growth

Data on trunk circumference were recorded at 10 cm above the graft union each year from 2014 to 2019 and were used to calculate Trunk Cross Sectional Area (TCSA). Canopy height and radius were recorded for each tree every year. Canopy volume was calculated from the formula $v = 2/3\ (\pi r^2 h)$, where v is the canopy volume, r is the mean canopy radius, and h is the canopy height [18].

2.4.2. Fruit Yield

Fruit yield for each experimental tree was recorded at each commercial harvest from 2015 to 2019, and Cumulative Yield (CY) was calculated for five years. Yield efficiency (YE) was calculated by dividing the CY by the TCSA of 2019 [19].

2.4.3. Fruit Size Distribution

Fruit from each tree was sorted into five size classes based on <65 mm (>138 fruit/carton), 65–68 mm (138–125 fruit/carton), 69–74 mm (113–100 fruit/carton), 75–77 mm (88 fruit/carton), and >77 mm (<80 fruit/carton) using a commercial grader [Colour Vision Systems Pty. Ltd., Melbourne, Australia (MAF Oceania group)]. These data were then used to determine fruit size distribution.

2.4.4. Alternate Bearing Index (ABI)

ABI was calculated using the deviation in fruit yield in successive years (*I*) from 2015 through to 2019 [20]:

$$\mathrm{ABI} = 1/\mathrm{n} - 1 * [(\,|\,\mathrm{a}_2 - \mathrm{a}_1\,|\,/\mathrm{a}_2 + \mathrm{a}_1) + (\,|\,\mathrm{a}_3 - \mathrm{a}_2\,|\,/\mathrm{a}_3 + \mathrm{a}_2) + \ldots + (\,|\,\mathrm{a}_\mathrm{n} - \mathrm{a}_{(\mathrm{n}-1)}\,|\,/\mathrm{a}_\mathrm{n} + \mathrm{a}_{(\mathrm{n}-1)})] \quad (1)$$

n = number of years, a_1, a_2, . . ., a_n, a_{n-1} = fruit yield in corresponding years.

An ABI of 1 indicates a very strong tendency towards alternate (also called biennial) bearing, and ABI close to zero indicates that season-to-season yield variability was random.

2.4.5. Internal Fruit Quality

The measurements for internal fruit quality were carried out at harvest for fruit from each experimental tree for five growing seasons. Ten fruit per tree were randomly selected at harvest from around the tree canopy and taken to the laboratory to assess internal fruit quality. Percent TSS of the juice was measured as °Brix with an Atago PAL-1 refractometer (www.atago.net Japan, accessed on 15 March 2024), and % citric acid (% acid) was estimated by titrating 10 mL of juice against standard 0.1 mol/L NaOH solutions. Juice sugar:acid ratio was calculated from this data [11]. BrimA was calculated as Brix − (acid × 4) × 16.5 [21].

2.5. Statistical Analyses

Statistical analyses were carried out using GenStat 21 [22], as a two-way analysis of variance with two factors (7 Valencia selections × 4 Trifoliata rootstocks), and mean separation was performed using Duncan Multiple Range test.

3. Results

3.1. Yield and Yield Components

There were significant differences in fruit yield each harvest season between trees on different rootstock selections. 'Zao Yang' had the highest yield with a cumulative yield increase of 27% (180 kg/tree) compared with the standard commercial rootstock 'Tri22' (137 kg/tree) (Table 2). Yield efficiency for 'Zao Yang' was 3.5 kg/TCSA compared with 3.1 kg/TCSA for 'Tri22'. 'Zao Yang' also displayed a low ABI of 0.19 as compared with other rootstocks, including 'Tri22' (Table 2). Cumulative yield data for each tree indicated that 'Valencia 5' trees had significantly greater yields (166 kg/tree) than other selections, except for 'Valencia 2' (156 kg/tree) (Table 2). 'Valencia 4' produced the lowest yield of 143 kg/tree compared to 'Valencia 5' (Table 2). The interaction effect between Rootstock × Scion was not statistically significant for cumulative yield; however, the 'Valencia 5'/'Zao Yang' combination produced 210 kg/tree, higher than 'Keenan'/'Tri22' and 'Valencia 4'/'Donghai' combinations, which each produced 127 kg/tree. The yield efficiency for 'Valencia 1' was higher than 'Keenan' and other selections, except 'Valencia 2'. The interaction effect was significant for yield efficiency (Figure 1). Yield efficiency was higher in 'Valencia 4'/'Zao Yang' and 'Valencia 1'/'Zao Yang' (3.8) compared with 'Keenan' (3.2). In this trial, 'Valencia 5'/'Donghai' had the lowest yield efficiency of 2.1.

Table 2. Yield components for seven Valencia selections on four Trifoliata rootstocks from 2015–2019 growing seasons.

	Fruit Yield (kg)/Tree					Cumulative Yield (kg)/Tree	Yield Efficiency (kg)/TCSA (cm^2)	Alternate Bearing Index (ABI)
Main Factors	**2015**	**2016**	**2017**	**2018**	**2019**	**2015–2019**	**2015–2019**	**2015–2019**
Rootstock								
Tri22 (control)	33 a	17 a	25 a	32 a	32 a	137 a	3.1 b	0.22 ab
Donghai	35 ab	19 a	26 a	32 a	33 a	144 a	2.9 a	0.22 bc
Tanghe	37 b	17 a	27 a	31 a	32 a	144 a	3.1 b	0.24 c
Zao Yang	41 c	26 b	31 b	43 b	36 b	180 b	3.5 c	0.19 a
Scion								
Keenan (control)	37 ab	21 c	24 b	40 b	32 ab	153 a	3.2 b	0.23 a
Valencia 1	32 a	20 bc	21 a	40 b	34 bc	147 a	3.4 c	0.20 a
Valencia 2	36 ab	24 d	30 c	30 a	36 c	156 ab	3.3 bc	0.22 a
Valencia 3	38 bc	19 abc	28 c	32 a	30 a	146 a	3.1 b	0.21 a
Valencia 4	33 ab	17 a	26 bc	34 a	32 ab	143 a	3.2 b	0.21 a
Valencia 5	42 c	20 abc	36 d	34 a	34 bc	166 b	3.0 a	0.24 a
Valencia 6	37 abc	18 ab	27 bc	32 a	34 bc	148 a	3.2 b	0.21 a
Analysis of variance								
Rootstock	***	***	***	***	**	***	***	***
Scion	***	***	***	***	***	**	***	NS
Rootstock × Scion	NS	NS	NS	NS	***	NS	***	NS

The probability value for the ANOVA, ** $p < 0.01$, and *** $p < 0.001$, and NS (non-significant). Mean separation within the columns was tested using Duncan's multiple range test ($p < 0.05$). Mean followed by the same letter within the column was not significantly different.

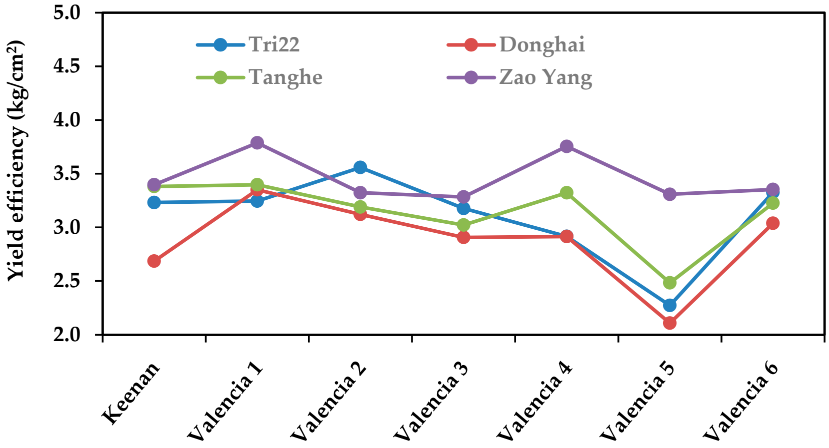

Figure 1. The interaction effect of Rootstock × Scion on yield efficiency (kg/cm^2) (2015–2019). The effect was significant at $p < 0.001$.

3.2. Fruit Size Distribution

Fruit size distribution data demonstrated significant main and interaction effects. Trees grafted to 'Zao Yang' rootstock had the highest percentage of fruit above 75 mm diameter (43%) compared with other rootstocks, and 'Valencia 6' had the highest percentage of large fruit (47%) compared with all other Valencia selections trialled. The 'Valencia 6'/'Zao Yang' combination had the highest percentage of large sized fruit (58%), significantly higher than fruit produced on the control trees of 'Keenan'/'Tri22' (35%). The lowest percentage of fruit greater than 75 mm diameter was produced by the 'Valencia 4'/'Tanghe' (19%) and 'Valencia 1'/'Tri22' (22%) combinations (Table 3).

Table 3. The percentage of large fruit (>75 mm) for seven Valencia selections budded to four Trifoliata rootstocks, averaged for the 2015–2019 growing seasons.

	Tri22 (Control)	Donghai	Tanghe	Zao Yang	Scion (Mean)
Keenan (control)	35 defgh	36 defg	39 fghij	44 hijk	38 c
Valencia 1	22 ab	29 bcde	25 abc	31 bcdefg	27 a
Valencia 2	37 efghi	34 cdefg	40 ghij	49 k	40 c
Valencia 3	31 bcdefg	39 fghij	26 abcd	38 efghi	34 b
Valencia 4	30 bcdef	27 abcd	19 a	48 jk	31 ab
Valencia 5	27 abcd	25 abc	33 cdefg	37 efghi	31 ab
Valencia 6	40 ghij	43 hijk	46 ijk	58 l	47 d
Rootstock (Mean)	32 a	33 a	32 a	43 b	

The main effect of Rootstock is significant at $p < 0.001$ and Scion at $p < 0.001$. The treatment differences were tested using Duncan's multiple range test ($p < 0.05$) and should be compared within the column for Scion and within the row for Rootstocks. The interaction effect of Rootstock × Scion was significant at $p < 0.001$ and should be compared across different rootstock and Scion combinations. Mean followed by the same letter within the column for (Scion) or within the row for (Rootstock was not significantly different. Means followed by the same letter for row and column combinations for the interaction effect (Rootstock × Scion) was not significantly different.

3.3. Fruit Quality Components

Data for TSS (measured as °Brix) varied across the growing seasons for the Valencia and rootstock selections. Generally, there was no effect of rootstock selection on TSS each year or on the average across the five years. However, fruit acidity was affected by rootstock treatment. Fruit produced on 'Zao Yang' rootstock had the highest acid value (0.98%) compared with other rootstocks (Table 4). Sugar:acid ratios, juice%, and BrimA values were not significantly affected by rootstock selections.

Scion variety had a significant effect on fruit quality over the five growing seasons. Fruit produced on 'Valencia 5' trees had significantly increased levels of TSS compared with 'Keenan' Valencia' each season. In the 2015, 2016, and 2018 seasons, fruit produced on

'Valencia 5′ trees had an average of 12.0 °Brix compared with 10.7 °Brix in fruit produced on 'Keenan' Valencia'. The average °Brix values across the 5 years were 1.7 degrees higher for 'Valencia 5′ than 'Keenan' Valencia' (Table 4). The average acid values and sugar:acid ratios across the five seasons were higher in fruit produced on 'Valencia 5′ trees than fruit produced on 'Keenan' and trees of the other selections. Juice percentage was slightly higher in 'Valencia 5′ compared to all other selections. The effects were more prominent in BrimA values across Valencia selections. 'Valencia 5′ had a BrimA value of 131 compared with 'Keenan' at 110 (Table 4). The interaction effect of Rootstock × Scion was not statistically significant for °Brix values; however, the 'Valencia 5′/'Zao Yang' and 'Valencia 5′/'Donghai' combinations produced 12.3 °Brix, compared with 10.0 °Brix for 'Keenan'/'Tri22′. The interaction effect of Rootstock × Scion was statistically significant for %acid values (Figure 2): the 'Valencia 5′/'Zao Yang' combination produced acid values of 1.1, compared with 0.9 for the 'Keenan'/'Tri22′ combination.

Table 4. Fruit quality variables total soluble solids (°Brix), acid (%), Brix:acid ratio, juice (%), and BrimA for 2015–2019 growing seasons.

	Total Soluble Solids (°Brix)					**°Brix**	**Acid (%)**	**°Brix:Acid Ratio**	**Juice (%)**	**BrimA**
Main Factors	**2015**	**2016**	**2017**	**2018**	**2019**	**2015–2019**	**2015–2019**	**2015–2019**	**2015–2019**	**2015–2019**
Rootstock										
Tri22 (control)	10.7 a	11.0 a	8.0 a	10.6 a	12.8 a	10.6 a	0.96 b	11.2 a	52 a	112 a
Donghai	10.6 a	11.0 a	7.9 a	10.3 a	12.7 a	10.5 a	0.94 a	11.2 a	51 a	111 a
Tanghe	10.9 b	11.0 a	7.8 a	10.7 a	12.8 a	10.6 a	0.95 ab	11.3 a	51 a	113 a
Zao Yang	10.9 b	11.1 a	7.8 a	10.6 a	12.9 a	10.7 a	0.98 c	11.1 a	51 a	111 a
Scion										
Keenan (control)	11.1 b	10.6 a	7.5 a	10.4 a	12.3 a	10.4 abc	0.94 a	11.3 bc	50 a	110 abc
Valencia 1	10.3 a	10.6 a	7.5 a	10.2 a	12.1 a	10.1 a	0.93 a	11.0 abc	50 a	106 a
Valencia 2	10.5 a	11.0 ab	7.9 a	10.2 a	12.8 ab	10.5 bc	0.93 a	11.3 c	51 a	111 bc
Valencia 3	10.5 ab	11.4 b	7.8 a	10.5 a	13.2 b	10.7 c	0.97 b	11.1 bc	52 a	112 c
Valencia 4	10.5 ab	10.8 ab	7.7 a	10.5 a	12.3 a	10.4 ab	0.97 b	10.8 a	51 a	107 ab
Valencia 5	12.0 c	12.2 c	9.3 b	11.9 b	14.8 c	12.1 d	1.03 c	11.9 d	53 b	131 d
Valencia 6	10.5 ab	10.4 a	7.3 a	9.9 a	12.2 a	10.1 a	0.93 a	11.0 ab	51 a	105 a
Analysis of variance										
Rootstock	*	NS	NS	NS	NS	NS	*	NS	NS	NS
Scion	***	***	***	***	***	***	***	***	**	***
Rootstock × Scion	NS	NS	NS	NS	NS	NS	**	NS	NS	NS

The probability value for the ANOVA, * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$, and NS (non-significant). Mean separation within the columns was tested using Duncan's multiple range test ($p < 0.05$). Mean followed by the same letter within the column was not significantly different.

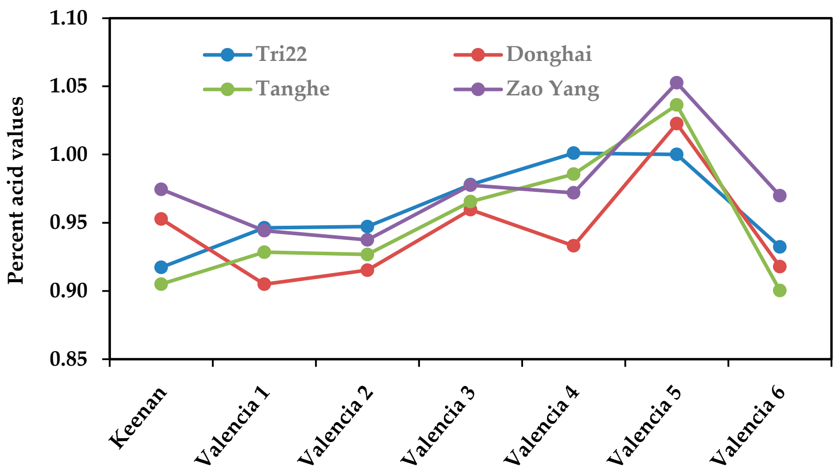

Figure 2. The interaction effect of Rootstock × Scion on mean percent acid values (2015–2019). The effect was significant $p < 0.01$.

3.4. Tree Growth Components

The trunk circumference measured during 2014 and 2019 was significantly greater for trees on 'Zao Yang' rootstock compared with 'Tri22' (Table 5). The rate of trunk circumference increment was higher for 'Zao Yang' and 'Donghai' compared with 'Tanghe' and 'Tri22'. For the scion treatments, 'Valencia 5' had significantly increased trunk circumference compared with 'Keenan' in both 2014 and 2019. The trunk circumference increments over the five seasons followed the same trend. Tree height differences across rootstocks became more significant over time as the trees grew. 'Zao Yang' and 'Donghai' trees were taller than trees on 'Tanghe' and 'Tri22' in 2019, and this difference was more evident in the height increment from 2014 to 2019 (Table 5).

Table 5. Tree growth variables trunk circumference (cm), tree height (m), canopy volume (m^3) and canopy spread (m) from 2014 to 2019 growing seasons.

	Trunk Circumference (cm)			Tree Height (m)			Canopy Volume (m^3)			Canopy Spread (m)		
Main Factors	**2014**	**2019**	**Increase**	**2014**	**2019**	**Increase**	**2014**	**2019**	**Increase**	**2014**	**2019**	**Increase**
Rootstock												
Tri22 (control)	14.7 a	23.8 a	9.1 a	1.47 ab	2.18 a	0.71 a	1.8 a	4.8 a	3.0 a	1.52 a	2.01a	0.50 a
Donghai	15.1 a	25.4 b	10.3 b	1.45 a	2.26 b	0.81 b	1.8 a	5.3 b	3.4 bc	1.54 ab	2.07 b	0.54 a
Tanghe	15.1 a	24.2 a	9.0 a	1.52 b	2.21 ab	0.69 a	2.0 b	5.2 b	3.2 ab	1.56 bc	2.08 bc	0.52 a
Zao Yang	15.6 b	25.9 b	10.2 b	1.51 b	2.34 c	0.82 b	2.0 b	5.7 c	3.7 c	1.59 c	2. 14 c	0.55 a
Scion												
Keenan (control)	15.2 c	24.7 b	9.5 a	1.46 ab	2.17 ab	0.71 a	1.9 b	4.7 b	2.8 ab	1.56 d	2.02 b	0.46 a
Valencia 1	14.1 a	23.2 a	9.1 a	1.39 a	2.07 a	0.68 a	1.5 a	4.0 a	2.5 a	1.44 a	1.92 a	0.47 ab
Valencia 2	15.0 bc	24.5 b	9.5 a	1.51 b	2.18 ab	0.67 a	1.9 b	4.7 b	2.8 ab	1.53 bcd	2.08 b	0.48 ab
Valencia 3	15.1 bc	24.3 b	9.1 a	1.50 b	2.22 b	0.73 a	1.9 b	4.9 b	3.1 b	1.55 cd	2.06 b	0.51 ab
Valencia 4	14.6 ab	23.7 ab	9.2 a	1.41 a	2.15 ab	0.74 a	1.7 a	4.7 b	3.0 b	1.49 abc	2.03 b	0.54 b
Valencia 5	17.4 d	29.4 c	12.0 b	1.71 c	2.76 c	1.05 b	2.9 c	9.1 c	6.2 c	1.80 e	2.50 c	0.70 c
Valencia 6	14.6 abc	24.0 ab	9.4 a	1.45 ab	2.18 ab	0.73 a	1.7 a	4.6 b	2.9 ab	1.48 ab	2.00 b	0.52 ab
Analysis of variance												
Rootstock	**	***	***	*	***	**	*	***	***	**	***	NS
Scion	***	***	***	***	***	***	***	***	***	***	***	***
Rootstock × Scion	***	**	***	NS	**	*	NS	*	NS	*	NS	NS

The probability value for the ANOVA, * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$, and NS (non-significant). Mean separation within the columns was tested using Duncan's multiple range test ($p < 0.05$). Mean followed by the same letter within the column was not significantly different.

'Valencia 5' trees were significantly taller and had a faster growth rate (1.05) in 2014 and 2019 compared with the commercial standard 'Keenan' Valencia (0.71) and the other scion selections (Table 5). The canopy volumes were very similar for all treatments in 2014, but by 2019, trees on 'Zao Yang' had a larger canopy volume (5.7) than 'Tri22' (4.8) (Table 5). Trees of 'Valencia 5' had significantly greater canopy volumes compared with 'Keenan' in both 2014 and 2019. This was also evident in canopy increment data that was highly significant for 'Valencia 5' (6.2) as compared to 'Tri22' (2.8). 'Valencia 1' trees had the lowest canopy volume increase of 2.5 compared with 'Valencia 3, 'Valencia 4', or Valencia 5' (Table 5). The interaction effect of Rootstock × Scion was significant for the circumference increase (Figure 3). 'Valencia 5'/'Zao Yang' combination had increased trunk circumference of 12.8 cm compared with 7.8 cm for 'Keenan'/'Tri22' combination. Tree heights and canopy volumes were also significantly influenced by the interaction between Rootstock × Scion treatments, except for 2014 (Table 5).

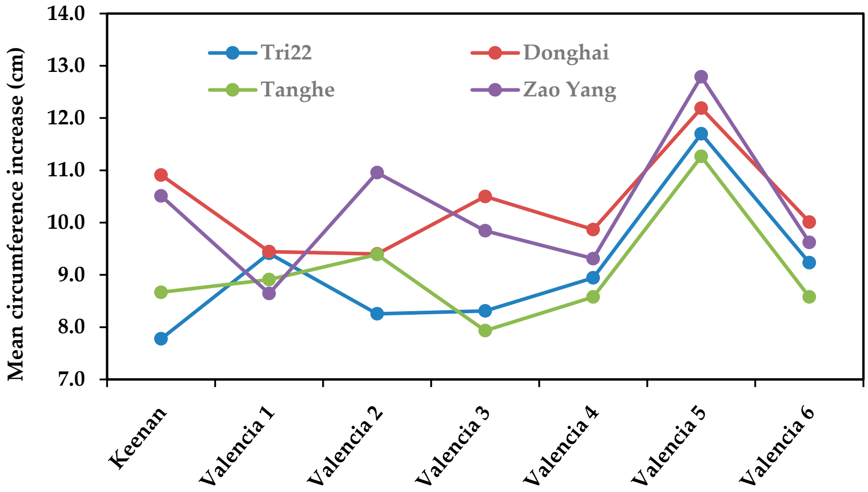

Figure 3. The interaction effect of Rootstock × Scion on mean circumference (cm) increase (2014–2019). The effect was significant $p < 0.001$.

4. Discussion

This study investigated the performance of 'Valencia' sweet orange selections on *P. trifoliata* rootstocks in a fully replicated field trial on a commercial orchard in the Riverina region of NSW. The new Trifoliata rootstocks under evaluation were released by NSW DPI due to their high performance in fully replicated field trials conducted at Dareton Primary Industries Institute under the deep sandy loan soil conditions [3]. The new experiment was designed to assess their performance in Australian main Valencia growing region, with a cooler climate and heavier textured soil. Although initially only available to Australian and Chinese citrus industries, these selected rootstocks will soon become available through Auscitrus to other citrus producing countries as public access seed.

The yield data collected in this trial has demonstrated the superior performance of 'Zao Yang' compared with 'Tri22', the standard selection used in clay (heavy) soil conditions in Australia. Our results for the yield data were consistent throughout five growing seasons. However, fruit yield varied across seasons. Annual yield variation in Valencia sweet orange is common due to the alternate bearing nature of this variety [23–26] and in 'Murcott' [27] and 'Nules' clementines [28]. Alternate bearing is caused by fruits inhibiting the formation of flowers on the same branch. The tendency of trees towards alternate bearing increases with an increase in age [29,30]. In this study, the trees were only nine years of age at the last harvest but were already exhibiting yield variations. Trees on 'Zao Yang' rootstock had a slightly lower alternate bearing index than the other rootstock varieties trialled. This result was encouraging as, typically, rootstock variety has no effect on alternate bearing [20,30]. Due to the variable yield across different growing seasons, it was more meaningful to assess the cumulative yields for five seasons. This showed that the cumulative yield for the 'Valencia 5' selection was the highest. However, the most striking feature of the data was that 'Valencia 5' trees on 'Zao Yang' had a 65% increase in fruit yield over the standard control combination of 'Keenan Valencia' on 'Tri22'.

Commercial citrus trees are pruned and hedged regularly affecting canopy volume. However, tree circumference typically expands each year. Therefore, the yield efficiency, calculated by measuring the trunk circumference, gives a reliable measure of the tree size compared with tree canopy volume [31]. In our work, trunk circumference explained 73% of the variation in the cumulative yield. 'Valencia 5' trees on 'Zao Yang' had 30% greater yield efficiency over the control combination. This was because the high yield induced by 'Zao Yang' and 'Valencia 5' scion more than compensated for their impacts on trunk growth. Furthermore, this result demonstrates that the superior performance of 'Valencia 5' is not a consequence of the source budwood being free of CTV, because 'Keenan' was also free of this virus, and yet its growth rate and productivity were similar to the five other scions [16].

Choice of rootstock can impact a wide range of variables [13]. The combination of rootstock and scion selection has shown to exert its effect on fruit quality of 'Or' and 'Odem' mandarins and Valencia oranges [32]. Fruit size is one of the important variables in sweet orange production, particularly where fruit are destined for fresh consumption. Therefore, it is significant that trees on 'Zao Yang' rootstock produced 43% of fruit above 75 mm in diameter, while the 'Valencia 6' scion selection had enhanced fruit size when compared with other Valencia selections. The combination of 'Valencia 6' on 'Zao Yang' is a good candidate to produce the larger fruit sizes that are often demanded by fresh fruit markets. Large sized fruit can generate high returns for citrus growers.

Internal quality variables are important for both juice processing and fresh fruit consumption [11]. In our study, 'Valencia 5' was found to be a superior scion because trees of this selection produced fruit with higher sugar levels than the other Valencia selections. 'Valencia 5' was able to consistently produce high sugar levels for five consecutive years, and this was more pronounced when 'Valencia 5' was grafted to 'Zao Yang' rootstock. The impact of scion and rootstock combination on fruit quality has been reported previously for mandarins [32]. Climatic conditions influenced the sugar and acid levels in the 2017 and 2019 growing seasons across all rootstock and Valencia selections but did not affect the overall trends exhibited in the quality variables. In the 2017 growing season, fruit had lower °Brix values due to an extremely cold winter and the number and extent of frost events that occurred between May and September (Table 1). An early freeze or drastic fluctuations in temperature has been found to exacerbate cold injury [33,34]. In 2019, very hot summer conditions and an irrigation system failure for several weeks in February caused the °Brix values to increase above expected values. Water stress imposed in late summer to early autumn in Australia has been reported to enhance sugar and acid levels at the expense of fruit size in Satsuma mandarins [35]. This has also been reported in early navel oranges in California [36]. Fruit yields were lower for 2019 in our studies, and this could likely be due to the reduced fruit size caused by heat and water stress. The consistent performance of 'Valencia 5' and 'Zao Yang' despite significant variations in seasonal climatic conditions further supports their commercial suitability. Generally, a set of 5 years of data is a reliable indicator for making recommendations, as suggested in [13].

The 'Valencia 5' selection also produced fruit with high acid levels and high sugar:acid ratios. Processing fruit with a blend of 12 °Brix and 1% acid is considered to produce good quality juice for the consumer (pers. comm. H. Baird, Manager Mildura Fruit Juices). It was also recommended that 12 °Brix and 1% acid to be ideal for producing good quality orange juice [37]. BrimA values are a Californian–Australian quality standard for harvest maturity [21], and it measures the balance between °Brix (sweetness) and acidity (sourness) [38]. The sugar:acid ratio is currently used as a maturity index for fruit. However, this measurement does not always correlate with perception of sweetness or sourness [39]. BrimA is a better indicator of flavour (taste) and was adopted in 2012 by the Californian Department of Food and Agriculture as the standard quality parameter to determine sweetness of Navel oranges [21]. BrimA is now also used by the Australian citrus industry to define fruit maturity. In our work, 'Valencia 5' produced higher BrimA values than any other selection. An additional advantage of 'Valencia 5' was its higher juice percent compared with all six other Valencia selections, which would make it more acceptable for the juicing industry.

'Valencia 5' is a clonal mutation selected in the Riverina as a single tree in a Valencia orchard. The 'Valencia 5' selection has been registered as 'DV' Valencia and has Plant Breeders Rights (PBR) protection. 'Zao Yang' trifoliate rootstock is commercially available to Australian growers through the Auscitrus budwood and seed scheme, with young trees now established in a range of soil types in southern Australia.

5. Conclusions

This study reports on the performance of seven Valencia scions on four Trifoliata rootstocks in Australia's main orange growing region, the Riverina of southern NSW. 'Valencia 5′ produced the highest cumulative yield and had significantly higher levels of TSS, being 1.7 degrees higher on average than the current industry standard 'Keenan'. Trees on 'Zao Yang' rootstock produced the highest yield, again outperforming the industry standard 'Tri22'. Therefore, the combination of 'Valencia 5′/'Zao Yang' is recommended for the juicing industry. 'Valencia 6′ on 'Zao Yang' rootstock had the highest percentage of fruit larger than 75 mm in diameter compared with other scion and rootstock combinations. Consequently, 'Valencia 6′/'Zao Yang' is recommended for fresh consumption due to its large fruit size and probably higher monetary returns. The results and recommendations from this evaluation trial have assisted the commercialisation of 'Valencia 5′ 'and 'Zao Yang' for sweet orange production in Australia, and potentially have importance for international rootstock and scion diversity.

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