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RESEARCH ARTICLE

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Effect of harvesting with a trunk shaker and an abscission chemical on fruit detachment and defoliation of citrus grown under Mediterranean conditions

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Abstract

Spain ranks as the world's leading exporter of citrus for fresh consumption. Manual harvest accounts for 50% of the total production costs. Mechanical harvest would increase labor productivity and benefits of growers. Efficiency of these machines depends on the varieties and operating conditions. Use of abscission chemicals has been promoted to increase the detachment rate of fruit without affecting its quality. This work is aimed at studying whether the mechanical harvest and/or the application of an abscission agent affect the quality and quantity of harvested fruit and tree defoliation under the conditions of citrus cultivation in Spain. Trials were made in a completely randomized experimental design. From 2008 to 2011, different orchards of mandarin and orange trees were sprayed with different doses of ethephon as abscission agent and harvested with a trunk shaker. Harvest related variables (detachment percentage, defoliation and fruit without calyx) were measured. The percentage of fruit detached by the trunk shaker ranged between 70 and 85% and it did not depend on the orchard. The shaker produced minimal damage to the bark when gripped incorrectly. Increased doses of ethephon increased fruit detachment except in 'Clemenules' orchard, but also increased the fruit without calyx in 1-9%. Moreover, ethephon promoted significant defoliation. Neither gummosis nor death of branches was observed. This work demonstrates that mechanical harvesting with trunk shakers may be a feasible solution for citrus cultivated in Spain for fresh market. Use of ethephon could only be recommended for citrus destined to industry and only for certain varieties.

Additional key words: mechanization; orange (*Citrus sinensis* (L.) Osb); clementine (*Citrus clementina* Hort. *ex* Tan.); ethephon; efficiency

Abbreviations used: CCI (citrus colour index); FRF (fruit retention force); LSD (least square difference); MI (maturity index); MLR (multiple linear regression); SE (standard error); TSS (total soluble solids); V_H (volume higher), V_L (volume lower), V_{Veg} (vegetation volume).

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Introduction

Spain is the leading exporter of fresh citrus with over 3 million tonnes per year (CLAM, 2010). The Valencian Region is the country's leading producer of citrus fruit – mainly mandarins – and grows more than 80% of the total national output (MARM, 2010). However, citrus production costs in Spain are higher than those of competitor countries, such as USA (Florida or California), Morocco, Egypt and Israel. Harvesting is performed manually and accounts for 29% of total direct production costs of oranges and 43% of mandarins in Andalucia (Junta de Andalucía, 2014a,b), in other regions it can be as much as 10 times higher than in competitor countries (Juste *et al*., 2000). Mechanisation of harvest would increase labour productivity and thus result in greater profits for agricultural entrepreneurs.

Totally or partially mechanised collection of fruit does take place with some crops in Spain, but not citrus. For instance, picking platforms are used with espaliered apple and pear groves, canopy shakers are employed for trellised vines and intensive olive groves, and limb and trunk shakers are used in extensive olive and almond groves.

The system to be used for harvesting the fruit depends largely on what the fruit is going to be used for. Fruit destined to the processing industry could be mechanically harvested because certain types of damage on the peel of the fruit are acceptable. In contrast, fruit destined to be eaten fresh cannot have any kind of damage, whether internal or external.

In Florida state (USA), where most of the production of citrus is used to make juice, mechanical harvesting has been widely studied over the last 50 years (Whitney, 1995). There, air shaker systems, trunk shakers and limb shakers or canopy shaker have all been tested (Sumner, 1973; Whitney & Wheaton, 1987; Whitney, 1997; Peterson, 1998; Ebel *et al*., 2010).

As reported in the literature, the efficiency of these machines depends on the fruit variety and the operating conditions. Li *et al*. (2005) obtained a fruit detachment rate of 90% in 'Hamlin' and 'Valencia' oranges with a trunk shaker vibrating at 4 Hz and with an amplitude of 13 cm applied for 10 s. Whitney *et al*. (2000a) achieved detachment rates of 85% in 'Valencia' oranges and between 57 and 71% in 'Hamlin' oranges with vibrations applied between 5 and 15 s. In recent years the first experiments have been conducted in Spain with oranges and mandarins using trunk shakers, resulting in detachment rates of between 57 and 77% (Torregrosa *et al*., 2009).

In an attempt to increase the performance of these machines, the use of abscission chemicals was promoted, above all in USA. Abscission chemicals, or agents, are exogenous plant-growth regulators that make it easier for the fruit to detach from the stalk in one of the abscission zones (stalk-calyx or calyx-fruit), with the aim of increasing the detachment rate, without affecting the quality of the product. The abscission agents that have been most widely studied are shown in Suppl. Table S1 [pdf online].

The application of abscission agents increased the percentage of detachment achieved by limb shakers by 20-35%. Thus, detachment percentages of 81-91% in 'Valencia' oranges and 93-100% in 'Hamlin' oranges were reported (Whitney *et al*., 1986; Whitney & Wheaton, 1987). Nevertheless, fruit destined to fresh market must be totally free of blemishes, as they lower its commercial value. At the same time, it must keep the calyx attached to the skin, since this is an indicator of fruit freshness and its absence could favour fungal growth.

No references have been found in the scientific literature about the application of abscission chemicals in the mechanized harvesting of citrus under Mediterranean climate conditions, which are drier and colder than those in Florida. Moreover cultural practices (irrigation, pruning, etc.) and grown cultivars are different. Despite the fact that no abscission chemicals have been legally registered on citrus in Spain, they appear to be interesting as an element that could help in harvesting with shakers and, as a result, lower harvesting costs. The only abscission agent that could be used in the short term is ethephon. Information on the effects of mechanised harvesting together with the application of ethephon in varieties of mandarins and oranges grown in the Mediterranean area is scarce in the scientific literature. Hence, the aim of this work was to determine the effectiveness of harvesting with trunk shakers, with or without the use of this abscission agent by analysing the results related to the percentage of detached fruit, the defoliation and the proportion of fruit that is harvested without calyx.

Material and methods

Description of the orchards. Treatments and dates

Eleven tests were conducted on five commercial orange (*Citrus sinensis* (L.) Osb) and mandarin groves, including clementines (*Citrus clementina* Hort. *ex* Tan.) and hybrids (*Citrus clementina* Hort. *ex* Tan. × *Citrus tangerina* Hort. *ex* Tan.) during the seasons 2008-09, 2009-10 and 2010-11. The characteristics of each grove are shown in Table 1. These groves produced from early ('Marisol') to late hybrid mandarins ('Fortune'), that are harvested in autumn and winter, thus covering the part of the harvest season in which there are few data from other countries.

Five treatments were carried out in each test: one control (water) and four different doses of ethephon (Ethrel 48, Numarf España, S.A., Barcelona, Spain) resulting from the combination of (i) two concentrations (600 and 1200 ppm) and (ii) two spray volumes, one higher, which was defined as the volume of liquid until the runoff point (V_H), and one lower, which was defined as a 40% reduction of the higher volume (V_L). These volumes varied according to the volume of vegetation in the canopy of the trees (V_{Veg}) of each variety and are shown in Table 2. An adjuvant (Mojante Inagra, Sipcam Inagra, S.A., Valencia, Spain) was added to the treatments at 0.05% to facilitate distribution of the product, as it has been indicated in the literature (Burns *et al*., 1999, 2006a,b; Kender *et al*., 2000; Pozo & Burns, 2009). The pH of the mixture was around 6.8-7

Table 1. Characteristics of the orchards: location, tree age, tree spacing and canopy volume.

Orchards	Location	Tree age (yr)	Tree spacing (m)	Canopy volume[a] (m^3/tree)	Observations and harvest time
Orogrande A	39º 35' 57" N 0º 22' 11" W	12-13	6 × 2	11.22	Mid-late mandarin season (October-January)
Orogrande B	39º 35' 56" N 0º 22' 14" W	12-13	6 × 2	11.22	Mid-late mandarin season (October-January)
Marisol	39º 39' 9.08" N 0º 18' 39.74" W	27	5 × 3.8	8.66	Early mandarin season (September-October)
Navel Lane Late	39º 36' 0.07" N 0º 21' 48.65" W	20	5.4 × 2.4	9.5	Mid orange season (January-May)
Clemenules	39º 28' 57.52" N 0º 36' 53.51" W	12	6 × 4	18.8	Mid-late mandarin season (November-January)
Fortune	39º 36' 49.22" N 0º 21' 12.79" W	22	5.8 × 4	18.8	Hybrid, late mandarin season (February-April)

[a]Canopy volume was calculated as the mean of three replicates considering citrus canopy as an ellipsoid with the tree dimensions of height, diameter 1 and diameter 2.

Table 2. Spray volume of treatments, meteorological data and operative characteristics of trunk shaker for each orchard and season.

Orchard	Season	Spray volumen of treatment		Meteorological data			Treatments date		Operative characteristics of trunk shaker		
		V_H (L/tree)	V_L (L/tree)	Mean T (ºC)	RH (%)	P (mm)	Spray	Days Spr-Vib	F (Hz)	A (mm)	t (s)
Orogrande A (mandarin)	2008-09	7	4	14.2	82	0.07	5/11/08	6	14.7	25	5+5
	2009-10			17.1	68	0.02	30/10/09	12	15.4	27	3+2
	2010-11			13.8	61	0.003	2/11/10	10	15.5	15	3+3
Orogrande B (mandarin)	2009-10	7	4	17.1	68	0.02	30/10/09	12	15.4	27	3+2
	2010-11			13.8	61	0.003	2/11/10	10	15.5	15	3+3
Marisol (mandarin)	2009-10	7	4	21.1	67	0.16	24/9/09	8-9	15	25	5
	2010-11			21.3	62	0.002	6/10/10	12	15	25	5
Navel Lane Late (orange)	2009-10	6	3.5	14.5	89	0.02	15/3/10	8	15.7	30	3+2
	2010-11			12.3	75	0.06	22/3/11	8	15.7	30	3+2
Clemenules (mandarin)	2009-10	8	4.5	13.7	59	0.002	23/11/09	10	14.7	17	3+2
Fortune (hybrid mandarin)	2009-10	10.5	6.5	13.2	71.	0.008	31/3/2010	7	14.1	35	3+2

V_H: volume higher; V_L: volume lower; T: temperature; RH: relative humidity; P: pluviometry; F: frequency; A: amplitude; t: time of vibration; Days Spr-Vib: days between spray treatment and vibration

in all the tests. Trials were made in a completely randomized experimental design. The experimental unit was one tree and each treatment was repeated five times, with a total of 25 trees per test.

The treatments were carried out with a hydraulic handgun sprayer, with a working pressure of 3 MPa and a cone angle of 30°. The higher water volume was applied using a ceramic conical nozzle of 1.2 mm diameter, whereas the lower water volume was applied by means of the same type of nozzle but with 1 mm diameter.

Between 6 and 12 days after applying ethephon (Table 2), all the trees were harvested with an orbital trunk shaker (Topavi, model: vibrator support arm, Maquinaria Garrido S.L., Autol, La Rioja, Spain) equipped with a three-point grip system (Fig. 1). Table 2 shows the operating characteristics (frequency, F; amplitude, A; and duration of vibration, t) of the shaker in each test. The frequency in the different tests ranged between 14.1 Hz and 15.5 Hz and the amplitude between 15-35 mm (Ortiz & Torregrosa, 2013). The duration of vibration was 5 s in all trials except for the first trial. Shaking was applied in one (5 s) or in two times (3 s+2 s), except in the first trial, in which trees were shacked 10 s (5 s +5 s). Previous work (Torregrosa *et al.,* 2009) demonstrated that these shaking

Figure 1. Trunk shaker used during the experiments. Left: trunk shaker with the shaker clamps open. Right: shaker attaching the mandarin trunk.

Table 3. Colour index, maturity index and fruit retention force (mean ± SE) at the harvest time depending on the variety and the season.

Orchard	Season	Harvest data (d/m/y)	Colour index	Maturity index	Fruit retention force (N)
Orogrande A	2008-09	11-12/11/2008	–8.97±0.48	11.00±0.17	70.79±1.19
	2009-10	9-10/11/2009	–9.14±0.61	10.52±0.20	68.92±2.64
	2010-11	11-12/11/2010	–13.00±0.30	12.98[a]	76.21±7.17
Orogrande B	2009-10	9-10/11/2009	–6.95±0.85	11.11±0.26	66.24±1.50
	2010-11	11-12/11/2010	–12.69±0.32	12.98[a]	71.86±2.00
Marisol	2009-10	2-3/10/2009	–13.86±0.55	7.17±0.27	46.35±1.09
	2010-11	18/10/2010	–17.26±0.39	8.16[a]	36.55±0.94
Navel Lane Late	2009-10	23/03/2010	10.31±0.48	12.97±0.32	123.50±2.66
	2010-11	30/03/2011	5.86±0.50	8.8[a]	146.50±3.85
Clemenules	2009-10	3/12/2009	3.22±0.43	13.02±0.19	70.47±1.76
Fortune	2009-10	7/04/2010	16.25±0.31	5.44±0.11	50.53±1.31

[a] Values provided by Fontestad S.A.

patterns have little importance in the results, since fruit and leaves are detached in the first 3 s. Frequency and duration of vibration were measured with a triaxial accelerometer placed on the tree trunk, near the shaker arm, registering the whole duration of the shakes with a digital oscilloscope at a frequency of 585 Hz. Amplitude was measured with video records at 300 frames per second. During shaking both fruit and leaves fell onto canvases that were arranged under each tree to catch them.

The dates when tests began were defined by the commercial demand for the fruit and the weather. Weather conditions (temperature, mean relative humidity and rainfall) from 15 days before the application until harvesting are shown in Table 3. Between the application of ethephon and the harvest with the trunk shaker there was very little rainfall. The small amount of rain fallen in the early days of the season before the 'Marisol' mandarins were harvested had no influence on the effect of ethephon, because the 8 h needed for the plant to absorb the product had already elapsed when the rain started (Wilson *et al*., 1977, 1981).

Description of the variables related with the state of the fruit on the harvesting dates

To determine the state of the fruit before applying ethephon, five fruits were picked at random from control trees, leaving a minimum stalk length of 2 cm. The fruit retention force (FRF) and citrus colour index (CCI) were measured for each fruit. Total soluble solids (TSS), acidity, and maturity index (MI) were measured for each juice extracted from the sample of five fruits of control trees. The FRF was measured using a digital dynamometer (Advanced Force Gauge 500 N, Mecmesin, England) by holding the stalk of the fruit horizontally in a fixed clamp and leaving 0.5 cm free until the calyx. The fruit was then pulled with the dynamometer, using a structure that allowed to pull the fruit horizontally. The colour of the peel was measured with a Minolta Colorimeter (Model CR-400/410; Japan) with the Hunter Lab coordinates (Jiménez-Cuesta *et al.*, 1981). The CCI of each fruit was calculated as the mean of two measurements taken on the equatorial zone, one in the green side and the other in the orange one. The MI was calculated as the ratio between the soluble solids and the acidity (González-Sicilia, 1968). The concentration of soluble solids was measured with a digital refractometer (Atago model PAL-3; Atago Co., Tokyo, Japan). Acidity was determined by titrating 5 mL-aliquots of juice with a 0.1 N solution of NaOH, with an automatic titrator (Mettler Toledo T50, Rondo Tower, Switzerland). In the case of the 'Marisol' and 'Fortune' mandarins, 3 mL-aliquots of juice were titrated because, being more acidic varieties, a greater amount of NaOH was needed.

Description of the harvest-related variables

Harvesting efficiency of the shaker was measured with the following two variables: fruit detached (%) and fruit detached without calyx (%). The variable defoliation was also measured. It was not possible to measure the percentage of defoliation because we were not allowed to defoliate the trees since trials were performed in commercial orchards, so we measured the amount of leaves (kg) detached per tree.

Fruits fallen after mechanical shake were weighed with a digital dynamometer (Advanced Force Gauge 500 N, Mecmesin, England). Fruits remaining on the tree after shaking were manually harvested and weighed. The relation between the amount of fruit detached with the shaker and the total amount of fruit on the tree (harvested with the shaker plus hand-picked) was used to calculate the percentage of fruit detached by the shaker.

The percentage of fruit without calyx was obtained from a random sample of 100 fruits detached by the shaker. All leaves detached from each tree after shaking were collected and weighed to evaluate defoliation.

Data analysis

First, the influence of the factors *Season* and *Orchard* on the efficiency of the trunk shaker was studied. This was carried out for the trees that were not treated with ethephon (dose 0) using multifactor analysis of variance on the data concerning percentage of fruit detached and percentage of fruit without calyx. Least Square Difference (LSD) test was used for mean comparisons. In this study, the assumption of normal distribution of data was assessed using the normal probability plot of the residuals and the assumption of homoscedasticity using the Levene's test (Levene, 1960). In all the analyses a confidence level of 95% was considered.

Second, the effect of ethephon dose on these same variables was studied, also including the influence of the factors *Orchard* and *Season*. Multiple Linear Regression (MLR) was performed to study the relationship between each of the two dependent variables (percentage of fruit detached and percentage of fruit detached without calyx) and the ethephon dose. In order to test whether these relationships were affected by the factors *Orchard* and *Season,* indicator variables were included in the regression model. An indicator variable is one that takes the value 0 or 1 to indicate the absence or presence of a categorical effect that may be expected to shift the outcome. When an indicator variable has n categories, only $(n - 1)$ indicator variables are introduced in order to avoid multicolinearity. The category for which the indicator is not assigned is known as the base group (Suits, 1957). In the present case, the factor *Orchard* had 6 categories ('Orogrande' A, 'Orogrande' B, 'Marisol', 'Navel Lane Late', 'Clemenules' and 'Fortune' orchards) and 'Orogrande' A orchard was chosen as the base group. The factor *Season* had 2 categories (2009-10 and 2010-11) and 2010-11 was chosen as the base group. MLR analysis followed an iterative process in which all the experimental data were included. It started by including the ethephon dose as independent variable, the two indicator variables (*Orchard* and *Season*) and their interactions in the model. Then the variable with the highest, non-significant p-value ($\alpha > 0.05$) was eliminated and the model was recalculated until all variables present in the model had significant coefficients. In all fitted models, all the assumptions of linear regression were checked. No outliers were identified.

Third, because the 'Orogrande' A orchard was the only one that was studied along the three seasons (2008-09, 2009-10 and 2010-11), the analysis of the effect of the season on the results of the different ethephon doses on this orchard was performed following the previous methodology. In this case, the factor *Season* had 3 categories (2008-09, 2009-10 and 2010-11) and 2010-11 was chosen as the base group.

Due to the fact that different orchards had different sizes and leave densities, the variable defoliation (kg leaves/tree) should not be compared among orchards. For this reason, the effect of ethephon dose was studied for each orchard. In the orchards studied along several seasons ('Orogrande A', 'Orogrande B', 'Marisol', 'Navel Lane Late'), the effect of the season was also studied. MLR was performed following the above methodology. As stated before, the factor *Season* had 2 or 3 categories depending of the orchard (2008-09, 2009-10 and 2010-11) and 2010-11 was chosen as the base group in all cases.

Results

State of maturity of the fruit at harvesting

Table 3 summarises the values (mean ± standard error) of the state of maturity of the fruit from the reference trees in each orchard and season at the time of harvesting (CCI, MI and FRF). In season 2010-11, fruit was greener than in the season 2009-10 in all orchards, since CCI were lower. The MI values did not vary much from one season to another for each orchard, except for 'Navel Lane Late' orchard which fruit had lower MI in season 2010-11 than in 2009-10. In general, MI values ranged between 11-13 in the less acid varieties ('Orogrande', 'Navel Lane Late' and 'Clemenules') and were lower (between 5 and 8) in the more acid ones ('Marisol' and 'Fortune'). In any case, all the CCI and MI values are considered adequate for marketing in our agroclimatic conditions.

The FRF values did not vary much from one season to another in each orchard. In all the seasons of 'Orogrande' and 'Clemenules' orchards, the FRF ranged between 66 and 76 N; in 'Marisol' and 'Fortune' orchards between 36 and 50 N; and in 'Navel Lane Late' orchard it was between 123 and 146 N.

Efficiency of the trunk shaker without applying ethephon

Percentage of fruit detached

The values of the detachment percentage for the different orchards and seasons tested are shown in Table 4 (dose 0). In season 2009-10, no significant differences were found in the detachment percentages between the orchards (F = 2.16; df = 5, 29; p = 0.0929), with values between 70 and 85%. In the season 2010-11, no significant differences were found in the detachment percentages between the orchards as well (F = 0.69; df = 3, 39; p = 0.5662), with values between 62-71%. However, significant differences were found among seasons, in 2009-10 significantly more fruit was detached than in 2010-11 (75% *vs* 67%, respectively) (F = 8.84; df = 1, 39; p = 0.0053).

When analysing the 'Orogrande' A orchard across three seasons (2008-09, 2009-10 and 2010-11), a non-significant reduction of the percentage of fruit detached was observed. In season 2008-09 the percentage of fruit detached was 78%, in season 2009-10 was 75% and in season 2010-11 dropped to 72%. This decrease may be due to the state of maturity of the fruit at the time of harvesting. In the first two seasons values of FRF, CCI and MI were similar (around 70 N for FRF, CCI = –9.00 and an MI of 12), however, in season

Table 4. Percentage of fruit detached (%) (mean ± SE) depending on the orchard, the season and the ethephon dose sprayed.

Orchard	Season	Ethephon dose sprayed (mg/tree)				
		0	**2400**	**4200**	**4800**	**8400**
Orogrande A	2008-09	78.46±3.84	71.80±4.09	78.31±4.24	75.61±5.18	81.93±3.59
	2009-10	74.98±5.46	78.60±2.98	78.12±3.86	84.42±2.30	84.47±3.60
	2010-11	71.76±3.22	67.02±4.24	66.25±7.17	70.67±3.65	72.59±4.49
Orogrande B	2009-10	82.01±0.80	87.97±2.14	89.55±1.02	87.04±1.18	93.79±1.49
	2010-11	62.81±2.87	71.77±3.08	74.11±3.42	78.41±3.11	84.47±1.95
Marisol	2009-10	72.98±4.66	77.96±3.65	79.47±3.72	90.18±2.67	93.30±2.17
	2010-11	66.03±6.08	74.69±1.99	79.44±3.93	78.86±6.46	80.58±4.62
		0	**2100**	**3600**	**4200**	**7200**
Navel Lane Late	2009-10	71.14±3.49	77.96±5.42	80.20±2.90	84.82±2.41	87.62±3.69
	2010-11	66.03±1.46	70.55±3.79	73.70±5.46	72.06±4.42	70.64±5.36
		0	**2700**	**4800**	**5400**	**9600**
Clemenules	2009-10	84.52±1.56	81.48±5.32	86.17±1.67	79.80±3.21	83.56±1.99
		0	**3900**	**6300**	**7800**	**12600**
Fortune	2009-10	70.29±5.74	67.48±2.97	80.70±2.39	73.00±2.37	73.48±2.98

2010-11 fruit was greener (CCI = –13.00, FRF = 76 N and MI = 12.98).

Percentage of fruit without calyx

The values of the percentage of fruit without calyx for the different orchards and seasons tested are shown in Table 5. In season 2009-10, significant differences of the percentage of fruit without calyx were found between orchards (F = 9.19; df = 4, 23; p = 0.0003). The percentage of fruit detached without calyx was higher in 'Fortune' orchard (9.3%), a little lower in 'Clemenules' orchard (6.2%) and much lower in the others: 'Navel Lane Late' orchard (3%) and 'Orogrande' A orchard and 'Orogrande' B orchard (1.3 and 0.8%, respectively).

In the orchards shaken in seasons 2009-10 and 2010-11 ('Navel Lane Late', 'Orogrande' A and 'Orogrande' B orchards) statistically significant interaction was observed between factors *Season* and *Orchard* (F = 8.56; df = 2, 28; p = 0.0017). For 'Navel Lane Late' orchard the percentage of fruit without calyx was very high in the season 2010-11 (8.4%) in comparison with the season 2009-10 (3%), however for 'Orogrande' orchards the percentage of fruit without calyx was similar between both seasons.

On the 'Orogrande' A orchard, in season 2008-09, percentage of fruit detached without calyx was significant higher (5%) than in the other two seasons (values around 1%) (F = 5.55; df = 2, 13; p = 0.0216).

Defoliation

Defoliation data are shown in Table 6 (dose 0). In the first season of mechanical harvest (season 2008-09 for 'Orogrande' A Orchard and season 2009-10 for the rest of orchards), defoliation was 0.74 kg leaves/tree in 'Marisol' 1.16 kg leaves/tree in 'Fortune', 1.45 kg leaves/tree in 'Orogrande' A, 1.69 kg leaves/tree in 'Orogrande' B, 1.79 kg leaves/tree in 'Navel Lane Late' and 1.86 kg leaves/tree in 'Clemenules'. It was visually estimated that these levels of defoliation represent between 3 and 6% of the total canopy.

In the orchards shaken in two consecutive years ('Marisol', 'Navel Lane Late', 'Orogrande' A and B), a drop in defoliation was observed in the second season in all orchards. The reduction of defoliation was 17.31% in 'Navel Lane Late', 22.06% in 'Orogrande A', 22.91% in 'Marisol' and 43.78% in 'Orogrande' B. In orchard 'Orogrande' A, defoliation of shaken trees in the third season was similar to that of the first season. Decrease of defoliation from the first to the second year of treatment may be due to the fact that in the first year the trees have a large number of senescent leaves that fall during shaking, whereas in the second year most of the leaves were young.

Effect of the ethephon dose

Percentage of fruit detached

The values of the detachment rate for the different orchards, seasons and doses of ethephon tested are

Table 5. Percentage of fruit without calyx (%) (mean ± SE) depending on the orchard, the season and the ethephon dose sprayed.

Orchard	Season	Ethephon dose sprayed (mg/tree)				
		0	**2400**	**4200**	**4800**	**8400**
Orogrande A	2008-09	5.42±1.59	13.53±2.34	16.43±0.53	24.79±2.76	28.37±4.67
	2009-10	1.30±0.78	4.04±2.28	6.45±2.66	3.79±0.87	14.24±5.98
	2010-11	1.36±0.50	4.00±2.07	5.80±1.11	8.40±0.93	12.45±3.82
Orogrande B	2009-10	0.80±0.49	17.30±4.16	27.84±2.38	32.74±6.53	43.62±4.22
	2010-11	0.60±0.24	4.39±1.91	5.91±0.95	6.40±2.25	15.20±2.96
Marisol	2009-10	ND	ND	ND	ND	ND
	2010-11	3.87±1.03	3.31±0.98	5.83±2.13	5.07±2.04	8.10±2.17
		0	**2100**	**3600**	**4200**	**7200**
Navel Lane Late	2009-10	3.00±0.71	17.55±1.64	35.06±7.18	60.96±5.92	70.68±5.31
	2010-11	8.42±1.56	11.00±4.73	25.15±8.84	28.25±6.25	36.87±8.59
		0	**2700**	**4800**	**5400**	**9600**
Clemenules	2009-10	6.15±1.47	6.20±3.07	10.83±2.70	12.68±3.80	17.39±4.33
		0	**3900**	**6300**	**7800**	**12600**
Fortune	2009-10	9.3±5.18	28.60±5.56	31.40±2.66	32.40±8.11	34.60±2.91

ND: no data.

Table 6. Defoliation (kg leaves/tree) (mean ± SE) depending on the orchard, the season and the ethephon dose sprayed.

Orchard	Season	Ethephon dose sprayed (mg/tree)				
		0	**2400**	**4200**	**4800**	**8400**
Orogrande A	2008-09	1.45±0.27	1.48±0.34	1.58±0.17	2.22±0.27	2.74±0.47
	2009-10	1.13±0.23	1.21±0.11	1.60±0.31	2.15±0.30	2.05±0.19
	2010-11	1.37±0.12	1.57±0.27	1.78±0.37	2.33±0.25	2.32±0.29
Orogrande B	2009-10	1.69±0.13	2.69±0.28	3.23±0.39	2.90±0.32	2.98±0.26
	2010-11	0.95±0.18	1.28±0.14	1.55±0.15	1.95±0.18	2.26±0.09
Marisol	2009-10	0.74±0.17	1.10±0.11	1.34±0.12	1.70±0.10	2.26±0.06
	2010-11	0.57±0.10	1.02±0.11	1.06±0.07	1.14±0.16	1.15±0.08
		0	**2100**	**3600**	**4200**	**7200**
Navel Lane Late	2009-10	1.79±0.21	3.27±0.25	4.20±0.54	5.13±0.33	7.18±0.17
	2010-11	1.48±0.17	3.19±0.63	2.84±0.35	4.46±0.40	5.35±0.83
		0	**2700**	**4800**	**5400**	**9600**
Clemenules	2009-10	1.86±0.05	2.61±0.17	3.19±0.18	3.35±0.16	3.70±0.20
		0	**3900**	**6300**	**7800**	**12600**
Fortune	2009-10	1.16±0.14	3.51±0.28	4.67±0.32	5.63±0.11	6.43±0.37

shown in Table 4. In general, when analysing season 2009-10, it can be seen that the rate of detachment increased significantly as the dose of the abscission agent increased. Such increase depended on the orchard, as shown by the fact that the indicator variables that multiplied the variable *Dose* were significant (except in 'Navel Lane Late' and in both 'Orogrande' orchards) (Table 7, row 1). It should be remembered that the significant indicator variables show changes of behaviour with respect to 'Orogrande' A orchard. The 'Marisol' orchard was more sensitive to the application of ethephon, since its indicator variable acted positively on the slope while 'Fortune' and 'Clemenules' orchards were less sensitive than 'Orogrande' A orchard (negative sign of the regression coefficient). 'Navel Lane Late' orchard and 'Orogrande' A orchard behaved in a similar manner (indicator variables associated to 'Navel Lane Late' orchard was not significant).

In the season 2010-11, again it can be seen that there was a direct effect of the dose of ethephon for the detachment rate ($F = 13.45$; df = 2, 98; $p < 0.0001$) and it differed from one orchard to another (Table 7, row 2). The response of the 'Orogrande' B orchard was more dose-sensitive than the others and was similar to that of 'Marisol' orchard. Like the previous year, 'Orogrande' A and 'Navel Lane Late' orchards were less sensitive.

On analysing the evolution of the data of 'Orogrande' A orchard over the three seasons considered in the study (2008-09, 2009-10 and 2010-11), significant variations were observed in the responses, since the regression coefficients that multiplied the indicator variables from the seasons 2008-09 and 2009-10 by the dose were significant. The trees were more sensitive to the dose in the season 2009-10 (Table 7, row 3).

Percentage of fruit without calyx

The values of the percentage of fruit detached without calyx for the different orchards, seasons and doses of ethephon tested are shown in Table 5. It can be seen that higher doses of ethephon resulted in higher percentages of fruit without calyx. Moreover, the percentage of fruit detached without calyx was higher in the first season of testing. On analysing the data by seasons, it can be observed that in the season 2009-10 the percentage of fruit detached without calyx due to the effect of ethephon differed significantly from one orchard to another (Table 7, row 4). The highest sensitivity to ethephon occurred in 'Navel Lane Late' orchard, followed by 'Orogrande' B orchard. The effect in 'Fortune' orchard was similar to 'Orogrande' A orchard and 'Clemenules' orchards, although there was less fruit without calyx in these latter cases.

Similar results appear in the season 2010-11. The 'Navel Lane Late' orchard was more sensitive to the effect of ethephon (Table 7, row 5) and the percentage of fruit without calyx was higher than the others orchards.

On analysing the evolution of this variable in 'Orogrande' A orchard over the three seasons studied, it was observed that the percentage of fruit detached without calyx was greater in the season 2008-09 and that in this same season the effect of dose was more pronounced (Table 7, row 6).

Table 7. Results of multiple linear regression analyses. D: Ethephon dose (mg/tree).

Regression analyses	Parameter[a]	Regression coefficient	T statistic	*p*-value
For the percentage of fruit detached in the season 2009-10 after ethephon treatment (F = 17.26; df = 6, 143; $p < 0.0001$). R^2 = 42.0007%.	Constant	73.3318	61.9266	<0.0001
	D	0.0017	6.1788	<0.0001
	(Orogrande B)	7.9710	4.5640	<0.0001
	(Clemenules)	10.0612	3.5947	0.0004
	D*(Fortune)	–0.0016	–5.5659	<0.0001
	D*(Clemenules)	–0.0017	–3.3014	0.0012
	D*(Marisol)	0.0007	1.9834	0.0492
For the percentage of fruit detached in the season 2010-11 after ethephon treatment (F = 13.45; df = 2, 98; $p<0.0001$). R^2 = 21.8882%.	Constant	69.2215	61.6188	<0.0001
	D*(Orogrande B)	0.0016	4.0353	0.0001
	D*(Marisol)	0.0016	4.0120	0.0001
For the percentage of fruit detached in the seasons 2008-09, 2009-10 and 2010-11 in 'Orogrande' orchard A after ethephon treatment (F = 8.73; df = 2, 72; $p = 0.0004$). R^2 = 19.9627%.	Constant	72.4549	53.2158	<0.0001
	D*(2008-09)	0.0010	2.3840	0.0198
	D*(2009-10)	0.0017	3.9859	0.0002
For the percentage of fruit without calyx in the season 2009-10 after ethephon treatment (F = 100.26; df = 4, 124; $p < 0.0001$). R^2 = 76.9685%.	Constant	2.6252	1.7335	0.0856
	D	0.0014	4.8167	<0.0001
	(Fortune)	16.9904	6.9659	<0.0001
	D*(Navel Lane Late)	0.0087	16.4031	<0.0001
	D*(Orogrande B)	0.0039	8.4607	<0.0001
For the percentage of fruit without calyx in the season 2010-11 after ethephon treatment (F = 28.03; df = 3, 74; $p < 0.0001$). R^2 = 54.2187%.	Constant	0.4419	0.1917	0.8485
	D	0.0015	3.1851	0.0022
	(Navel Lane Late)	10.3710	2.5892	0.0117
	D*(Navel Lane Late)	0.0021	2.2683	0.0264
For the percentage of fruit without calyx in the seasons 2008-09, 2009-10 and 2010-11in 'Orogrande' orchard A after ethephon treatment (F = 43.60; df = 3, 73; $p < 0.0001$). R^2 = 65.1393%.	Constant	0.6122	0.4252	0.6720
	D	0.0014	4.7253	<0.0001
	(2008-09)	6.1632	2.3646	0.0208
	D*(2008-09)	0.0014	2.5749	0.0121
For the defoliation in the three years of assays in 'Orogrande' orchard A after ethephon treatment (F = 29.26; df = 1, 73; $p < 0.0001$). R^2 = 28.8953%.	Constant	1.2424	9.8565	<0.0001
	D	0.0001	5.4092	<0.0001
For the defoliation in the two years of assays in 'Orogrande' orchard B after ethephon treatment (F = 40.15; df = 2, 49; $p < 0.0001$). R^2 = 63.0818%.	Constant	2.0897	13.4271	<0.0001
	D	0.0002	5.4836	<0.0001
	(2010-11)	–1.0988	–7.0879	<0.0001
For the defoliation in the two years of assays in 'Marisol' orchard after ethephon treatment (F = 45.31; df = 2, 49; $p < 0.0001$). R^2 = 65.8479%.	Constant	0.9392	10.7673	<0.0001
	D	0.0001	8.0107	<0.0001
	(2010-11)	–0.4468	–5.1426	<0.0001
For the defoliation in the two years of assays in 'Navel Lane Late' orchard after ethephon treatment (F = 61.31; df = 2, 49; $p < 0.0001$). R^2 = 72.29%.	Constant	2.1209	7.3514	<0.0001
	D	0.0006	10.6690	<0.0001
	(2010-11)	–0.8488	–2.9645	0.0047
For the defoliation in the assay in 'Clemenules' orchard after ethephon treatment (F = 60.52; df = 1, 24; $p < 0.0001$). R^2 = 72.46%.	Constant	2.0728	15.1521	<0.0001
	D	0.0002	7.7793	<0.0001
For the defoliation in the assay in 'Fortune' orchard after ethephon treatment (F = 141.28; df = 1, 24; $p < 0.0001$). R^2 = 85.99%.	Constant	0.264973	6.3304	<0.0001
	D	0.0000357614	11.8862	<0.0001

[a] In brackets significant indicator variables associated to the corresponding season or orchard.

Defoliation

The defoliation values (mean ± SE) for the different orchards, seasons and doses of ethephon tested are shown in Table 6.

In all orchards and seasons, the increase in ethephon dose produces a significant increase in defoliation (Table 7, rows 7-12). In the first year of testing (season 2009-10 for all the orchards except for 'Orogrande' A, which began in 2008-09), at the maximum dose the defoliation ranged between 6 and 7 kg leaves/tree in 'Navel Lane Late' and 'Fortune' orchards and 2-4 kg leaves/tree in the others orchards. It was visually estimated that defoliation was around 5-20% of total leaves. In the second year of experiments (season 2010-11 for all the orchards except 'Orogrande' A, for which the second was 2009-10), again it was observed that higher doses of ethephon produced significantly more defoliation. That season, 5.35 kg leaves/tree were shed in the 'Navel Lane Late' orchard with the highest dose and about 2 kg leaves/tree were detached in the others orchards. In general, once more, it can be seen how less defoliation took place in the second year of experimentation.

Data showed that the amount of leaves detached was not affected by the season and increased significantly with dose during the three years of testing in 'Orogrande' A orchard (Table 7, row 7).

Discussion

The percentage of fruit detached by the effect of the trunk shaker alone (without ethephon) ranged between 70 and 85% and, according to the statistical analysis, it did not depend on the orchard. The differences of these values could be due to the maturity of the fruit in each season.

These percentages could be improved if the trees were adapted to the mechanical harvest with the trunk shaker by adequate pruning. During the experiments it was observed that presence of flexible, long, thin, almost horizontal branches reduced the percentages of fruit detachment, since the vibration was damped. In a few cases the shaker was gripped incorrectly and caused slight bark scrapping. Similar experiences were reported in Florida (USA) (Li & Syversten, 2004, 2005). Several branches were also broken in the lower part of the canopy as a result of the manoeuvring required to enable the machine to reach the trunk, but if citrus trees had been adequately pruned this problem would not have happen.

The application of ethephon increased fruit detachment as the dose increased, except in 'Clemenules' orchard. The highest dose of ethephon increased the efficiency of the trunk shaker by 21% in 'Marisol' orchard, by 17% in 'Navel Lane Late' orchard, between 9-12% in 'Orogrande' orchards and by only 4% in 'Fortune' orchard. These values are similar to those obtained in Florida for early and late oranges (mainly 'Hamlin' and 'Valencia') where the percentage of fruit harvested was seen to increase by 5-15% (Koo *et al*., 1999; Whitney *et al*., 2000a,b; BenSalem *et al*., 2001; Farooq *et al*., 2002; Whitney, 2003). It is important to note that this did not occur in lemons grown under similar conditions (Torregrosa *et al*., 2010). Ethephon dose had little or no effect on shaker efficiency on 'Clemenules' and 'Fortune' varieties. One reason for this can be a specific balance of vegetal hormones in the fruit abscission zone, as Yuan *et al.* (2001a) pointed out in a study of factors affecting the physiological response of 'Valencia' oranges to abscission agents. Another reason may be the different weather conditions prior, during and after the applications of ethephon, as suggested by Yuan & Burns (2004). Despite the increased percentage of detached fruit, the doses that were applied also increased the percentage of fruit without calyx. This makes it more difficult for the product to be marketed in fresh, although it does not affect that destined to the juice industry or for the use in other industrial applications. In addition, it should be noted that the percentage of fruit detached without calyx by the effect of the trunk shaker alone ranged between orchards with values around 1-9%. However, the percentage of fruit without calyx was lower in the 2010-11 than in the 2009-10 season, when the fruit was riper. This could indicate that the greener the fruit the higher proportion of fruit with calyx can be harvested.

It was observed that defoliation caused by mechanical harvest was higher in the first year. This may be attributed to the fact that senescent leaves were removed this season and subsequently leaves in the following year were younger, therefore being more attached to the branches. The use of ethephon promoted significant defoliation as occurred with other abscission agents (Rasmussen, 1977; Hartmond *et al*., 2000a,b; Burns, 2002; Burns *et al*., 2003a,b; Pozo & Burns, 2006; Li *et al*., 2008). Indeed, defoliation increased as dose increased. However, despite this loss of leaves, the capacity of the trees to intercept light may not be severely affected (Li *et al*., 2006) because trees may partially compensate defoliation by increasing the capacity for photosynthesis of the leaves that remain in the canopy (Syversten, 1994).

It should be noted that after the applications of ethephon, no cases of gummosis or death of any branches were observed, as it happened in Florida after the application of other abscission agents like prosulfuron and metsulfuron-methyl (Whitney, 2003).

As a conclusion of this work, authors consider that mechanical harvest with trunk shakers could be a feasible solution for citrus cultivated in Spain destined to the fresh market, since high percentage of fruit detachment can be achieved, most of the fruit preserve their calyx and defoliation of the canopy is negligible. Use of ethephon as an abscission agent to increase the performance of mechanical harvest could only be recommended for citrus destined to the juice industry and only for the varieties which are affected by this chemical agent, like 'Marisol', 'Navel Lane Late' and 'Orogrande', but not for 'Clemenules' and 'Fortune'. In addition, the following research is envisaged. First, a study of an important collateral consequence of ethephon applications, which is their effect on peel colour changes and to asses if this has an influence on the commercial maturity of the fruit. Moreover, it is important to assess the short and long term effects of this abscissor and/or the shaker on the tree physiological status and yield. It is also considered that a specific study of the effect of ethephon applications on different plant organs and citrus varieties may be necessary. And last, but not least, authors recognize that the orchard indicator variable includes several features (variety, location, tree age, canopy volume, leaf area index, planting density, pruning level, cultural practices, etc.). It is known that these variables may have an effect on shaker efficiency and ethephon impacts and would require further work in the next future.

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