

Factors affecting phenology of different *Citrus* varieties under the temperate climate conditions of Santa Fe, Argentina

Norma Guadalupe Micheloud¹, Damián César Castro²,
Marcela Alejandra Buyatti³, Paola Maricel Gabriel⁴,
Norberto Francisco Gariglio⁵

ABSTRACT - The aim of this study was to characterize the phenology of different sweet orange, tangerines and tangerine hybrid varieties growing under the temperate climate conditions of Santa Fe Province, Argentina. Phenological stages were observed weekly during five consecutive years using a BBCH (Biologische Bundesanstalt, Bundessortenamt and Chemical industry) scale adapted for *Citrus* trees. All varieties showed a winter rest period from June to August. 'New Hall' and 'Navelina' varieties were the first to reach sprouting stage, whereas 'Okitsu' was the last. Inception of flowering occurred from August 13th to September 6th; and full bloom from September 12th to October 2nd. Fruit harvest started with the 'Okitsu' cultivar in March, and continued over a 7-month period. Interannual variation for inception of sprouting was high (44 days), and sprouting was correlated with both thermal accumulation (above 13°C) and the amount of solar radiation measured during July ($p < 0.0001$; $r^2 = 0.79$). Navel oranges and the 'Murcott' hybrid bloomed 5–15 days earlier than other varieties, increasing probability of damage by late frosts.

Index terms: BBCH scale, flowering, fruit set, harvest, sprouting, thermal accumulation.

Fatores que afetam a fenologia de diferentes variedades de citrus em clima temperado de Santa Fé, Argentina

RESUMO - O objetivo deste trabalho foi caracterizar o comportamento fenológico das diferentes variedades de laranja-doce, tangerinas e híbridos de tangerina que crescem em condições de clima temperado. Os estádios fenológicos foram observados semanalmente, durante cinco anos consecutivos, usando uma escala BBCH (Federal Biological Research Centre for Agriculture and Forestry) adaptada para citros. Todas as variedades apresentaram um período de repouso invernal entre junho e agosto. As primeiras variedades no início de brotação foram 'New Hall' e 'Navelina', e a última foi 'Okitsu'. O início da floração ocorreu no período de 13 de agosto a 6 de setembro, e do dia 12 de setembro até 2 de outubro observou-se a plena floração. A colheita dos frutos foi iniciada em março com a cv. 'Okitsu' e foi estendida durante sete meses com as outras variedades. A variação interanual na época do início da brotação foi elevada (44 dias), e seu tempo de ocorrência foi correlacionado com o acúmulo térmico (acima 13°C) e a quantidade de radiação solar durante julho ($p < 0.0001$; $r^2 = 0.79$). As laranjeiras-de-umbigo e o híbrido 'Murcott' apresentaram uma antecipação na floração de 5 a 15 dias, o que pode aumentar a probabilidade de danos ocasionados por geadas tardias.

Termos para indexação: BBCH escala, brotação, floração, frutificação, acumulação térmica, colheita.

Corresponding author:

E-mail: nmicheloud@fca.unl.edu.ar

Received : November 01, 2016

Accepted : May 04, 2017.

Copyright: All the contents of this journal, except where otherwise noted, is licensed under a Creative Commons Attribution License.



¹Agricultural Engineer, M.Sc. in Intensive Crops, Professor of Plant Physiology. Facultad de Ciencias Agrarias, Universidad Nacional del Litoral. Argentina. Kreder 2805, S3080HOF, Esperanza, Santa Fe Argentina. Telephone: +54 (3496) 426400. E-mail: nmicheloud@fca.unl.edu.ar

²Dr. Professor of Fruitculture. Facultad de Ciencias Agrarias, Universidad Nacional del Litoral. Argentina. E-mail: dcastro@fca.unl.edu.ar

³Agricultural Engineer, M.Sc. in Intensive Crops. Professor of Floriculture. Facultad de Ciencias Agrarias, Universidad Nacional del Litoral. Argentina. E-mail: mbuyatti@fca.unl.edu.ar

⁴Agricultural Engineer, Professor of Intensive Crops. Facultad de Ciencias Agrarias, Universidad Nacional del Litoral. Argentina. E-mail: pgabriel@fca.unl.edu.ar

⁵Dr. Professor of Fruitculture. Facultad de Ciencias Agrarias, Universidad Nacional del Litoral. Argentina. E-mail: ngarigli@fca.unl.edu.ar

Introduction

Citrus are native to the tropical and subtropical regions of Asia and the Malay Archipelago, and their commercial production is concentrated between 20°–40° latitude in both hemispheres (ROOSE et al., 2015). *Citrus* have higher value than other fruits in terms of international trade, and main *Citrus* producing countries include China, Brazil, the United States, Spain, Mexico, Italy, Egypt, Turkey, South Africa, and Argentina (FAO, 2012).

Perennial fruit production is an emerging economic activity in the central area of Santa Fe Province, Argentina (MICHELOUD et al., 2016; WEBER et al., 2013), which has been introduced over the past 10 years as an alternative for diversifying traditional production systems based on annual horticultural crops. *Citrus*, low-chilling peach and apple, fig, and raspberry crops are grown by marginal and small farmers (0.5–2 ha), who obtain good profitability through regional marketing (TRAVADELO et al., 2012).

Plant development is a progressive series of orderly, coherent changes leading to maturity (PEER et al., 2015). Development involves changes in functional ability and is greatly affected by genotype–environment interactions (PEREIRA et al., 2002). In perennial crops such as *Citrus*, the description of seasonal phenological stages is crucial to understanding crop function, and long-term phenological data are an important agronomic tool for crop management decisions and for climatic zone characterization (BARBASSO et al., 2005; OLIVEIRA et al., 2017; WEBER et al., 2013). Such records are used to predict phenological behavior of different varieties in a specific region by extrapolating experimental results obtained from other regions. Furthermore, phenological data can also be used to help predict interannual changes in tree phenology (PETRI et al., 2008). Thus, knowledge of the timing of phenological events and their variability can help producers obtain more stable crop yields and quality, thereby reducing environmental risks (GARIGLIO et al., 2012; SIMÕES et al., 2015).

Citrus species can be commercially cultivated both in temperate and subtropical environments. In temperate areas, annual cycles of plant growth are determined by the seasonal temperature changes (GRAVINA, 2007; MARTÍNEZ-FUENTES et al., 2013; MICHELOUD et al., 2016), whereas in tropical regions, they are also affected by rainfall distribution (MEDINA-URRUTIA et al., 2007; RIBEIRO and MACHADO, 2007). Consequently, knowledge of *Citrus* phenology in both environments can help producers and researchers obtain a better understanding of plant physiology and plant response to the environment.

Some phenological stages have a more pronounced effect on crop yield and are therefore called critical stages (AGUSTÍ et al., 1997; BARBASSO et al., 2005; OLIVEIRA et al., 2017). For example, some orchardist practices, such as pruning, fertilization, girdling, fruit

thinning, hormonal treatment, and pest and disease control, can be performed only at a given phenological stage, pointing to the need to identify growth stages accurately (ANDERSON et al., 2012; FAVARO et al., 2014; RIVAS et al., 2010). Phenological stages are described using phenological scales; the BBCH scale is the most widely accepted for *Citrus* (AGUSTÍ et al., 1997). This scale was simplified for growers in the Uruguay River region (Argentina) by Anderson et al. (2012).

In temperate zones, the timing of sprouting and flowering is critical for *Citrus* crops, because late frost occurrence can greatly reduce annual fruit yield (MICHELOUD et al., 2016). Furthermore, low winter temperatures increase *Citrus* flowering intensity (RIBEIRO et al., 2006; SOUTHWICK and DAVENPORT, 1986), which affects fruit set and fruit yield in some varieties (IGLESIAS et al., 2007). In the central area of Santa Fe, the average date for the last late-season frost is September 15th, coinciding with the beginning of *Citrus* flowering (GARCÍA et al., 2013). The average annual temperature in central Santa Fe is 19°C, and the average temperatures of the warmest and coldest months are 24.7°C and 11.5°C, respectively (GARCÍA et al., 2013).

The aim of this study was to generate records of the timing of phenological stages for different orange and tangerine varieties grown in the temperate climate conditions that define central Santa Fe Province, Argentina. We hypothesized that by increasing our knowledge of *Citrus* phenology and by identifying the main environmental factors affecting when flowering occurs, we could greatly reduce the risks inherent in *Citrus* production in central Santa Fe.

Materials And Methods

Plant material and growth conditions

Field trials were conducted in an experimental orchard and at a commercial farm, both located in Esperanza City, Santa Fe Province, Argentina (60° 50' W, 31° 25' S). The climate of this region is classified as type *Cfa*, which corresponds to a humid, temperate mesothermal climate (KÖPPEN, 1948).

Seven-year-old *Citrus* trees grafted onto *Poncirus trifoliata* (L.) Raf. rootstock, planted 5m × 3m apart in deep loamy (typical Argiudol) soil were used. Plants were provided with supplemental drip irrigation and chemical fertilizers according to their needs (PILATTI et al., 2009). Eight orange cultivars, all derived from *Citrus sinensis* (L.) Osbeck, were evaluated in this experiment: 'New Hall', 'Navelina', 'Washington navel', and 'Lane late' from the navel orange group; and 'Salustiana', 'Midnight', 'Delta seedless', and 'Valencia late' from the white orange group. Tangerine cultivars included were: 'Okitsu' (*Citrus unshiu* Marc.), 'Clemenules' (*Citrus clementine* Hort. Ex Tanaka), and *Citrus sinensis* (L.) Osb. x *C. reticulata* Bl. hybrids 'Ellendale' and 'Murcott'.

Treatments

During 5 consecutive years (2008–2012), phenological stages for each cultivar were recorded weekly using the Biologische Bundesanstalt, Bundessortenamt und Chemische Industrie (BBCH) scale adapted by Anderson et al. (2012). Vegetative stages from winter's end to full leaf development, and reproductive stages from the beginning of flowering to harvest, were recorded for each variety. The earliest and last dates for each phenological stage in the 5-year period were recorded and expressed in days. Meteorological data for the 5 years of study were obtained from a LI-COR 1400 automated weather station installed at the experimental orchard (Table 1).

Experimental design

We used a completely randomized, single-tree plot design with five replications per variety. Trees of each variety were selected based on their uniformity (size and vigor), and four branches, one from each of the four quadrants of the canopy, were selected from each tree (MARTÍNEZ-FUENTES et al., 2013) for observation. Data were tested for normal distribution and homogeneity of variance, and means were compared by the Least Significant Difference (LSD) test. Relationships between meteorological data and tree phenology were determined using InfoStat software (DI RIENZO et al., 2012). The best regression model was selected using the following criteria: minimizing the conditional model estimator (CME), the Akaike information criterion (AIC), and the Bayesian information criterion (BIC).

Results and Discussion

The mean monthly temperature from June–August was below or slightly above 13°C (Table 1). This value (13°C) is the threshold temperature for *Citrus* plant growth (SYVERTSEN and LLOYD, 1994; PIMENTEL et al., 2007), below which vegetative rest is induced. This rest condition has also been reported in other temperate and subtropical *Citrus* production areas around the world (GRAVINA, 2007; RIBEIRO and MACHADO, 2007).

The trees evaluated in our study started vegetative growth in August (Table 2) once the temperature exceeded the base threshold temperature for *Citrus* consistently (13°C; Table 1). The navel orange 'New Hall' was the first to reach sprouting stage (B1), whereas the 'Okitsu' tangerine cultivar was the last, 20 days later (Table 2). Averaged across the 5 years of study (2008–2012), other varieties sprouted from August 18th to the 28th. The full-leaf development stage (B6) finished in the third week of October or the first week of November, depending on variety, nearly 2 months after the commencement of sprouting (Table 2). Time elapsed from the beginning of sprouting to the full-leaf development stage (60 days) was consistent with a previously published report describing this phenological stage in 'Washington navel' oranges and

'Tahiti limes' (LOVATT and KRUEGER, 2015).

During the 5 years of our study, the phenological stage with the highest interannual variation was inception of sprouting (B1), at nearly 44 days. Interannual variation was 31 days at two other stages, elongated shoots with leaves unfolded and leaves not yet at full size (B3–4). At the full-leaf development stage (B6), interannual variation was only 19 days, which was less than half the variance observable at the B1 sprouting stage (44 days' interannual variance). Our results were consistent with two previously published studies on different early peach varieties grown in the same region (GARIGLIO et al., 2009; 2012). Results could be attributed to greater interannual variations in mean monthly temperature at the beginning of the growing season (Table 1), and to large increases in the monthly value of heat accumulation coinciding with advancement in the growing cycle. Faster thermal accumulation may have compensated for year-to-year phenological differences in a lower number of days, thus reducing phenological variability.

A significant relationship was found to exist for onset of sprouting with two variables: thermal accumulation (over 13°C) and solar radiation accumulation during July (Table 3). The combination of both variables (thermal accumulation and solar radiation) proved to be the best option to predict the onset of *Citrus* sprouting in central Santa Fe ($R^2=0.79$; Table 3). In contrast, no correlation was found between either temperature or radiation with sprouting during the months of May, June, or August (Table 3).

Previously published research shows that under low temperature conditions, especially under low soil or substrate temperatures, *Citrus* plants' metabolism is severely reduced; low night temperatures cause stomatal closure (via a decrease in stomatal conductance) and reduce net daily photosynthesis (via a decrease in biochemical reaction rates of carboxylation and ribulose-1,5-bisphosphate [RuBP] regeneration) (RIBEIRO et al., 2009; SANTOS et al., 2011). In addition, stomatal conductance reduction also increases leaf temperature in relation to air temperature (RIBEIRO et al., 2009), because plant transpiration is reduced (YU et al., 2015).

Based on previously published literature, we expected that the increase in leaf temperature of our *Citrus* plants would be directly proportional with solar radiation (MEDINA et al., 2002). Consequently, our calculation of plant heat accumulation based on air temperature underestimated the actual plant heat accumulation because air temperature was lower than leaf temperature (YU et al., 2015). This aspect was relevant because the median temperature in July was very close to the threshold temperature for *Citrus* metabolism (–1.3°C) (Table 1). Therefore, small differences between July temperatures and solar radiation across the 5 years could have had a large effect on metabolism and *Citrus* tree phenology. Furthermore, this effect may explain why inception of sprouting was better correlated with solar radiation

accumulation than with heat accumulation (based on air temperature) (Table 3). Although the prediction model we developed in our present study (Table 3) is validated for only the central Santa Fe region, it can be used to contribute to a broader understanding of *Citrus* phenology, especially in other regions with similar climates.

Root activity decreases markedly during the winter rest period, leading to a decrease in the synthesis and transport of plant hormones (e.g., gibberellin) and nitrogen compounds to new shoots (SOUTHWICK and DAVENPORT, 1986). Gibberellins are considered as strong inhibitors of flower induction (MUÑOZ-FAMBUENA et al., 2012), which explains why low temperatures act as the primary flower-inducing factor in *Citrus* (RIBEIRO et al., 2006). *Citrus* trees in our study (Table 4) and in other temperate *Citrus* production areas (GRAVINA, 2007; MARTÍNEZ-FUENTES et al., 2013; RIVAS et al., 2010) only develop flowers during the spring growth flush, and consequently, their annual fruit production is determined in this period. In contrast, several flowering cycles have been reported throughout the annual growing cycle under tropical climate conditions (RIBEIRO and MACHADO, 2007; RIBEIRO et al., 2006).

In our present study, inception of flowering (F1) occurred from August 13th through September 6th (Table 4), simultaneous with sprouting (Table 2). Full bloom (F4) was recorded from September 12th to October 2nd, depending on variety (Table 4). Furthermore, navel oranges had the highest flowering intensity, with 266 and 290 flowers per 100 nodes in 'Lane late' and 'Washington navel' varieties, respectively, in agreement with previously recorded results (GRAVINA, 2007; MICHELOUD et al., 2016; PILATTI et al., 2009). In *Citrus* trees, flower intensity affects the type of inflorescence and the time and rate of flower and fruit abscission (LOVATT and KRUEGER, 2015; RIVAS et al., 2010). In our current research, flower abscission occurred earlier after high flowering intensity; abscission began during full bloom and continued through to petal abscission stages (F4 and F5; Table 5).

The relative proportion of leafless inflorescences also increased with flowering intensity (IGLESIAS et al., 2007). Usually, leafless inflorescences emerge first and have a low probability of setting fruit, whereas flowers in leafy inflorescences emerge later and normally show higher fruit set (IGLESIAS et al., 2007; MARTÍNEZ-FUENTES et al., 2013). The positive influence of leaves on fruit set appears to be associated to net CO₂ assimilation and supply from developing leaves (LOVATT and KRUEGER, 2015). In the present study, this relationship could explain the early occurrence of fruit abscission at the F1 and F2 stages in navel oranges (Table 5). High flowering intensity could also explain reduced fruit set (0.1%) and the poor fruit yield we obtained from these orange varieties (MICHELOUD et al., 2016; PILATTI et al., 2009). The tangerines 'Okitsu' and 'Clemenules', and the orange 'Delta seedless', also showed early fruit abscission in 2009. This was probably due to the

tangerines' high proportion of leafless inflorescences and to the 'Delta seedless' orange's high flowering intensity (data not shown).

Fruit set (F7 stage) occurred from October 19th to 25th, depending on variety (Table 4). The precise identification of this growth stage is very important, because numerous agronomic practices, such as the application of exogenous gibberellins (GRAVINA, 2007; MARTÍNEZ-FUENTES et al., 2013) or girdling of branches (RIVAS et al., 2010) are performed at this time to improve fruit set and productivity.

The development of *Citrus* fruits occurs over a relatively long period that includes three phases (SYVERTSEN and LLOYD, 1994; IGLESIAS et al., 2007). In our present study, physiological fruit drop ended in late December (corresponding to June drop in the Northern Hemisphere) for all varieties (Table 6). This phase overlapped the end of the first stage of fruit development (IGLESIAS et al., 2007). Maturation was reached in the autumn in early varieties such as 'Okitsu', 'Clemenules', 'New Hall', and 'Navelina'; and in the winter in intermediate varieties such as 'Washington navel', 'Salustiana', 'Lane late', and 'Ellendale'. Finally, the later-maturing varieties such as 'Murcott', 'Midnight', 'Delta seedless' and 'Valencia late' reached maturity at the beginning of the spring (Table 6).

The maximum time between start of harvest for any two varieties with successive maturity was just 4 weeks. Maximum interval for start of harvest across all varieties used in our trial occurred between 'Delta seedless' and 'Valencia late' cultivars (Table 7). *Citrus* fruits respond to a non-climacteric maturation mechanism; ethylene production and respiration rate are low, and changes in texture and composition occur gradually in ripe fruits (IGLESIAS et al., 2007). These characteristics of *Citrus* fruits allow the fruits to remain on the plant for a variable period of time, and consequently, harvest period becomes continuous from March with the early tangerine 'Okitsu' to late September, when the 'Valencia late' orange is picked (Table 7).

Table 1. Radiation, air and soil temperature, rainfall, relative humidity, and potential evapotranspiration data recorded in central Santa Fe Province, Argentina.

		J	F	M	A	M	J	J	A	S	O	N	D
SR (Mj/m ²)	M	25.1	21.1	20.6	14.2	8.6	7.0	8.3	11.3	14.4	18.6	23.0	23.8
	CV (%)	7.7	6.9	27.2	19.8	12.4	14.1	22.2	10.9	6.8	9.1	12.5	7.2
Tmax (°C)	M	33.2	30.5	28.9	26.1	22.0	17.9	18.3	20.1	23.0	25.6	30.5	31.2
	CV (%)	3.4	2.5	2.6	4.4	6.1	5.9	10.9	16.3	8.1	5.6	3.3	5.8
Tmean (°C)	M	26.9	24.5	22.6	19.4	16.3	11.8	11.7	13.6	16.3	19.6	24.6	25.5
	CV (%)	4.2	5.1	4.2	2.8	7.6	8.4	19.7	18.3	9.8	4.2	3.2	4.0
Tmin (°C)	M	20.0	18.6	16.5	13.4	11.0	6.2	5.8	7.4	9.9	13.5	17.6	19.0
	CV (%)	6.2	7.3	5.8	6.4	10.7	26.0	47.8	30.1	12.6	10.5	7.6	4.8
ST (°C)	M	27.7	25.6	23.6	20.2	16.4	12.5	11.6	13.5	16.6	19.7	24.6	25.6
	CV (%)	5.6	6.1	5.7	4.5	7.1	7.8	16.6	11.7	4.6	1.5	1.5	5.3
AccP (mm)	M	201.2	194.2	175.2	79.0	47.4	9.4	20.6	36.6	116.4	165.0	183.2	201.1
	CV (%)	90.8	34.2	31.3	44.5	33.4	144.2	152.5	137.9	115.3	82.3	89.4	95.9
RH (%)	M	70.6	77.4	78.4	78.6	83.3	77.3	70.7	69.1	68.8	72.0	69.3	68.6
	CV (%)	11.2	5.4	5.3	8.2	7.9	8.1	7.5	12.9	14.4	10.9	9.1	11.05
ETp (mm)	M	136.2	116.6	111.6	85.1	67.1	48.1	50.6	62.4	80.7	105.0	134.1	147.8
	CV (%)	37.4	7.9	12.2	11.2	17.4	8.5	19.3	18.2	7.7	4.2	1.6	3.7

Note: Data are the means of a 5-year period (2008–2012).

Abbreviations: AccP, accumulated precipitation; CV, coefficient of variation; ETp, potential evapotranspiration; M, monthly mean value; RH, relative humidity; SR, solar radiation; ST, soil temperature; Tmax, maximum temperature; Tmean, mean temperature; Tmin, minimum temperature.

Table 2. Average dates for inception of vegetative growth stages for different *Citrus* varieties cultivated in central Santa Fe Province, Argentina.

Variety	B1	B2	B3	B3-4	B4	B5	B6
'New Hall'	13/08 ± 12	25/08 ± 14	30/08 ± 15	12/09 ± 5	20/09 ± 9	06/10 ± 7	21/10 ± 4
'Navelina'	18/08 ± 8	26/08 ± 8	05/09 ± 15	13/09 ± 11	23/09 ± 11	15/10 ± 5	26/10 ± 6
'Lane late'	20/08 ± 15	01/09 ± 16	12/09 ± 15	23/09 ± 11	05/10 ± 10	20/10 ± 9	04/11 ± 4
'Washington navel'	23/08 ± 13	04/09 ± 13	16/09 ± 14	26/09 ± 12	05/10 ± 13	19/10 ± 10	04/11 ± 6
'Delta seedless'	23/08 ± 13	01/09 ± 14	19/09 ± 8	27/09 ± 11	04/10 ± 8	20/10 ± 10	31/10 ± 6
'Valencia late'	23/08 ± 15	02/09 ± 13	12/09 ± 13	25/09 ± 12	05/10 ± 10	20/10 ± 11	01/11 ± 7
'Midnight'	27/08 ± 12	02/09 ± 13	16/09 ± 14	23/09 ± 11	03/10 ± 11	21/10 ± 11	05/11 ± 5
'Salustiana'	27/08 ± 11	02/09 ± 10	13/09 ± 9	02/10 ± 8	10/10 ± 12	20/10 ± 13	05/11 ± 5
'Murcott'	23/08 ± 10	02/09 ± 11	13/09 ± 12	02/10 ± 6	17/10 ± 7	23/10 ± 10	01/11 ± 5
'Ellendale'	27/08 ± 17	08/09 ± 14	16/09 ± 14	24/09 ± 13	02/10 ± 11	18/10 ± 6	01/11 ± 4
'Clemenules'	28/08 ± 13	06/09 ± 13	16/09 ± 13	23/09 ± 12	04/10 ± 8	20/10 ± 8	04/11 ± 4
'Okitsu'	02/09 ± 12	14/09 ± 11	24/09 ± 13	30/09 ± 7	09/10 ± 10	22/10 ± 12	05/11 ± 5

Notes: B1, inception of sprouting; B2, elongating shoots; B3, elongating shoots with leaves growing; B3-4, elongated shoots with leaves unfolded, not yet at full size; B4, shoots reaching their full size; B5, shoots and leaves mature; and B6, full-leaf development. Data and standard deviations (±) are the means of a 5-year period (2008–2012).

Table 3. Relationships of temperature (*T*) and solar radiation (*R*) variables with inception of sprouting (*y*, in July days) for different *Citrus* varieties cultivated in central Santa Fe Province, Argentina.

Model	R ²	<i>p</i> (model)
$y = 250.80 - 0.19 \times TT_{Jul}$	0.42	<0.0001
$y = 246.28 - 0.06 \times TT_{Aug}$	0.08	0.0845
$y = 201.97 + 0.15 \times R_{Jul}$	0.68	<0.0001
$y = 217.86 + 0.07 \times R_{Aug}$	0.12	0.0307
$y = 214.60 - 0.11 \times TT_{Jul} + 0.13 \times R_{Jul}$	0.79	<0.0001

Notes: Thermal accumulation (base temperature = 13°C) during July (*TTJul*) and August (*TTAug*); solar radiation (MJ.m⁻²) accumulated during July (*RJul*) and August (*RAug*). Data are the means of 5 years (2008–2012).

Table 4. Average dates for the reproductive growth stages of different *Citrus* varieties in central Santa Fe Province, Argentina.

Variety	F1	F2	F3	F4	F5	F6	F7
'New Hall'	13/08 ± 12	31/08 ± 15	07/09 ± 14	12/09 ± 8	20/09 ± 9	03/10 ± 10	19/10 ± 11
'Navelina'	18/08 ± 8	29/08 ± 12	06/09 ± 11	14/09 ± 12	21/09 ± 9	05/10 ± 13	23/10 ± 9
'Lane late'	20/08 ± 15	10/09 ± 17	20/09 ± 16	24/09 ± 14	30/09 ± 16	10/10 ± 10	23/10 ± 14
'Washington navel'	23/08 ± 13	09/09 ± 15	18/09 ± 14	23/09 ± 15	01/10 ± 15	11/10 ± 14	23/10 ± 14
'Delta seedless'	28/08 ± 15	10/09 ± 15	17/09 ± 13	21/09 ± 12	30/09 ± 14	11/10 ± 11	24/10 ± 13
'Valencia late'	01/09 ± 12	14/09 ± 12	22/09 ± 10	26/09 ± 10	05/10 ± 9	11/10 ± 7	23/10 ± 9
'Midknight'	28/08 ± 14	10/09 ± 14	20/09 ± 12	25/09 ± 14	03/10 ± 14	12/10 ± 11	25/10 ± 11
'Salustiana'	02/09 ± 11	17/09 ± 11	25/09 ± 9	29/09 ± 10	04/10 ± 9	12/10 ± 8	23/10 ± 8
'Murcott'	23/08 ± 10	09/09 ± 10	16/09 ± 11	19/09 ± 20	27/09 ± 13	04/10 ± 9	19/10 ± 9
'Ellendale'	29/08 ± 14	11/09 ± 12	20/09 ± 10	23/09 ± 20	27/09 ± 11	07/10 ± 15	17/10 ± 15
'Clemenules'	31/08 ± 12	17/09 ± 12	24/09 ± 14	27/09 ± 14	03/10 ± 12	12/10 ± 10	16/10 ± 14
'Okitsu'	06/09 ± 11	20/09 ± 12	30/09 ± 13	02/10 ± 13	09/10 ± 13	15/10 ± 13	23/10 ± 11

Notes: F1, onset of flowering (green buds); F2, flowers with petals closed; F3, first flowers open; F4, full bloom; F5, petal abscission; F6, petals fallen off; and F7, fruit set. Data and standard deviations (±) are the means of a 5-year period (2008–2012).

Table 5. Inception of reproductive structure abscission in different *Citrus* varieties grown in central Santa Fe Province, Argentina.

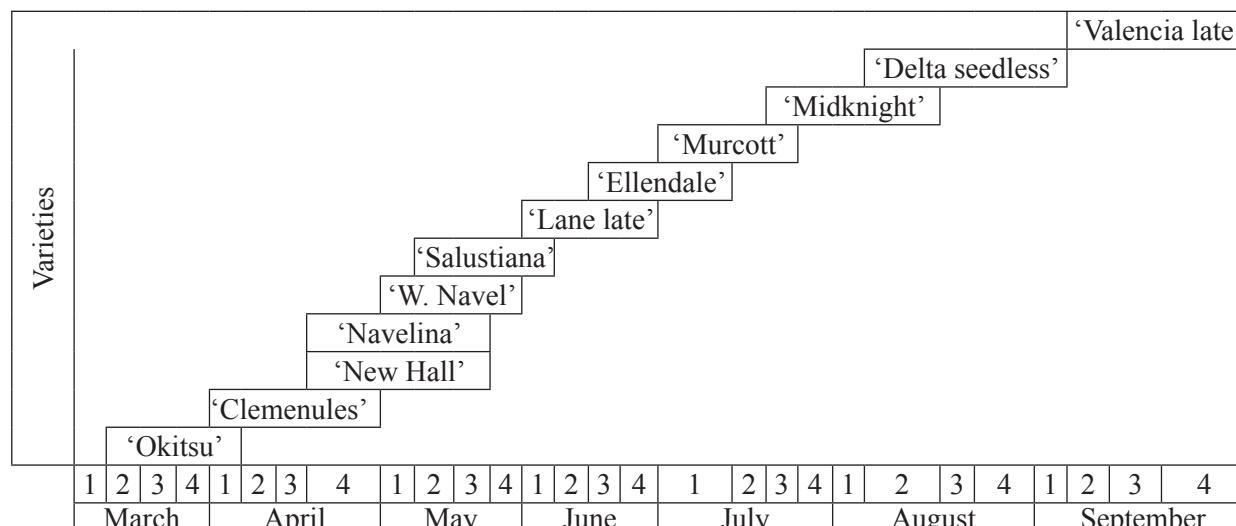
	2008	2009	2010	2011	2012
'Washington navel'	F2	F4	F4	F1	F3
'Lane late'	F2	F3	F5	F2	F4
'Delta seedless'	F5	F2	F4	F1	F4
'Midknight'	F3	F3	F3	F2	F4
'Ellendale'	F3	F5	F5	n.d.	n.d.
'Clemenules'	F4	F2	F4	F3	F4
'Valencia late'	F3	F4	F4	F5	F2
'Salustiana'	F3	F5	F4	F5	F3
'Okitsu'	F3	F2	F4	F2	F3

Notes: F1, onset of flowering (green buds); F2, flowers with petals closed; F3, first flowers open; F4, full bloom; and F5, petal abscission. Data are representative of 5 years' (2008–2012) growth. n.d.: No data.

Table 6. Phases of fruit development for different *Citrus* varieties grown in central Santa Fe Province, Argentina.

Variety	Phase I	Phase II	Phase III
'Washington navel'	23/09 – 14/11	14/11 – 05/04	05/04 – 20/05
'Lane late'	24/09 – 16/11	16/11 – 20/04	20/04 – 15/07
'Delta seedless'	21/09 – 15/11	15/11 – 23/02	23/02 – 20/08
'Midknight'	25/09 – 25/11	25/11 – 05/04	05/04 – 16/08
'Ellendale'	23/09 – 07/12	07/12 – 20/04	20/04 – 20/08
'Clemenules'	27/09 – 05/12	05/12 – 16/03	16/03 – 20/04
'Valencia late'	26/09 – 07/12	07/12 – 20/04	20/04 – 20/08
'Salustiana'	29/09 – 20/11	20/11 – 16/03	16/03 – 20/05
'Okitsu'	02/10 – 17/11	17/11 – 15/02	15/02 – 15/03

Notes: Phase I, cell multiplication; phase II, cell elongation; and phase III, maturation. Data are the means of 5 years (2008–2012).

Table 7. Harvest period of different *Citrus* varieties grown in central Santa Fe Province, Argentina.

Data are the means of 5 years (2008–2012).

Conclusions

Citrus grown in the central area of Santa Fe had a pronounced winter rest period from June to August, and a concentrated flowering period in the spring. The large interannual variation in onset of sprouting we observed in the current study can be explained by the combination of two variables, thermal accumulation and solar radiation accumulated during July ($R^2=0.79$). Navel oranges and the 'Murcott' hybrid bloomed earlier (by 5–15 days), which is a characteristic that increases the probability of damage by late spring frosts. Navel oranges also showed high flowering intensity that can affect fruit set and yield.

Acknowledgments

This study was supported by the National University of Littoral (grant CAID+D 2016, PI: 50120150100020LI). We also thank M.Sc. Rubén Andrés Pilatti for his assistance with the experiments and his critical review of the article.

References

AGUSTÍ, M.; ZARAGOZA, S.; BLEIHOLDER, H.; BUHR, L.; HACK, H.; KLOSE, R.; STAUB, R. Adaptation de l'échelle BBCH à la description des stades phénologiques des agrumes du genre *Citrus*. **Fruits**, Montpellier, v.52, n.5, p.287-295, 1997.

ANDERSON, C.M.; BANFI, G.; CASAFUS, C.M.; COSTA, N.B.; FABIANI, A.; GARRÁN, S.M.; MARCO, G.; MIKA, R.H. **Manual para productores de naranja y mandarina de la Región del Río Uruguay**. Concordia: INTA, 2012. p.203.

BARBASSO, D.V.; PEDRO JÚNIOR, M.J.; PIO, R.M. Caracterização fenológica de variedades do tipo Murcott em três porta-enxertos. **Revista Brasileira de Fruticultura**, Jaboticabal, v.27, n.3, p.399-403, 2005.

DI RIENZO, J.A.; CASANOVES, F.; BALZARINI, M.G.; GONZALEZ, L.; TABLADA, M.; ROBLEDO, C.W. **InfoStat versión 2012**. Córdoba: Universidad Nacional de Córdoba, Argentina, 2012. Disponible em: <<http://www.infostat.com.ar>>.

FAO. **Frutos cítricos frescos y elaborados**. Estadísticas anuales, 2012. Disponible em: <<http://www.fao.org/economic/est/est-commodities/citricos/es/>>.

FAVARO, M.A.; MICHELOUD, N.G.; ROESCHLIN, R.; CHIESA, M.; CASTAGNARO, A.; VOJNOV, A.; GMITTER, F.; GADEA VACAS, J.; RISTA, L.; GARIGLIO, N.F.; MARANO, M.R. Surface barriers of mandarin cv. 'Okitsu' leaves make a major contribution to canker disease resistance. **Phytopathology**, St. Paul, v.104, n.9, p.970-976, 2014.

- GARCÍA, M.S.; MICHELOUD, N.G.; LEVA, P.; TÓFFOLI, G.; GARIGLIO, N.F.; PILATTI, R.A. Efecto de las heladas tardías en la producción de durazneros precoces cultivados en la región central de la provincia de Santa Fe (Argentina). **Revista FAVE Sección Ciencias Agrarias**, Esperanza, v.12, n.1, p.135-143, 2013.
- GARIGLIO, N.F.; MENDOW, M.; WEBER, M.; FAVARO, M.A.; GONZÁLEZ-ROSSIA, D.; PILATTI, R.A. Phenology and reproductive traits of peaches and nectarines in central-east Argentina. **Scientia Agrícola**, Piracicaba, v.66, n.6, p.757-763, 2009.
- GARIGLIO, N.F.; WEBER, M.E.; CASTRO, D.; MICHELOUD, N.G. Influence of the Environmental Conditions, the Variety, and Different Cultural Practices on the Phenology of Peach in the Central Area of Santa Fe (Argentina). In: XIAOYANG, Z. **Phenology and climate change**. Croatia: InTech, 2012. p.217-240.
- GRAVINA, A. Aplicación del ácido giberélico en *Citrus*: revisión de resultados experimentales en Uruguay. **Agrociencia**, Montevideo, v.11, n.1, p.57-66, 2007.
- IGLESIAS, D.J.; CERCOS, M.; COLMENERO FLORES, J.M.; NARANJO, M.; RÍOS, G.; CARRERA, E.; RUIZ-RIBERO, O.; LLISO, I.; MORILLON, R.; TADEO, F.R.; TALON, M. Physiology of *Citrus* fruiting. Review. **Brazilian Journal of Plant Physiology**, Campinas, v.19, n.4, p.333-362, 2007.
- KÖPPEN, W. **Climatología**: un estudio de los climas de la tierra. Traducción Pedro H.R. Pérez. México: Fondo de Cultura Económica, 1948.
- LOVATT, C.J.; KRUEGER, R.R. Morphological and yield characteristics of 'Washington' navel orange and 'Tahiti' lime trees produced with buds from "floral" versus "vegetative" mother shoots. **Acta Horticulturae**, The Hague, v.3, n.1065, p.1831-1837, 2015.
- MARTÍNEZ-FUENTES, A.; MESEJO, C.; MUÑOZ FAMBUENA, N.; REIG, C.; GONZALES MAS, M.C.; IGLESIAS, D.J.; PRIMO MILLO, E.; AGUSTÍ, M. Fruit load restricts the flowering promotion effect of paclobutrazol in alternate bearing *Citrus* spp. **Scientia Horticulturae**, Amsterdam, v.151, p.122-127, 2013.
- MEDINA, C.L.; SOUZA, R.P.; MACHADO, E.C.; RIBEIRO, R.B.; SILVA, J.A.B. Photosynthetic response of *Citrus* grown under reflective aluminized polypropylene shading nets. **Scientia Horticulturae**, Amsterdam, v.96, p.115-125, 2002.
- MEDINA-URRUTIA, V.M.; ZAPIAIN ESPARZA, G.; ROBLEZ GONZALES, M.M.; PEREZ ZAMORA, O.; OROZCO SANTOS, M.; WILLIAMS, T.; BECERRA RODRIGUEZ, S. Fenología, eficiencia productiva y calidad de fruta de cultivares de naranjo en el trópico seco de México. **Fitotecnia Mexicana**, México, v.30, n.2, p.133-143, 2007.
- MICHELOUD, N.G.; CASTRO, D.C.; FAVARO, M.A.; BUYATTI, M.A.; PILATTI, R.A.; GARIGLIO, N.F. Response of some *Citrus* species to frost damage at the central area of Santa Fe, Argentina. **Revista FCA UNCUYO**, Mendoza, v.48, n.2, p.43-56, 2016.
- MUÑOZ-FAMBUENA, N.; MESEJO, C.; GONZALES-MAS, M.C.; IGLESIAS, D.; PRIMO-MILLO, E.; AGUSTÍ, M. Gibberellic Acid Reduces Flowering Intensity in Sweet Orange [*Citrus sinensis* (L.) Osbeck] by Repressing CiFTGene Expression. **Plant Growth Regulation**, Berlin, v.31, n.4, p.529-536, 2012.
- OLIVEIRA, C.R.M.; MELLO-FARIAS, P.C.; DE OLIVEIRA, D.S.C.; CHAVES, A.L.S.; HERTER, F.G. Water availability effect on gas exchanges and on phenology of 'Cabula' orange. **Acta Horticulturae**, The Hague, v.1, n.1150, p.133-138, 2017.
- PEER, W.; BEVERIDGE, C.; BUSOV, V.; MURPHY, A.; TAIZ, L. Vegetative growth and organogenesis. In: TAIZ, L.; ZEIGER, E.; MOLLER, I.M.; MURPHY, A. **Plant physiology and development**. 6th ed. Sunderland: Sinauer Associates, 2015. p. 553-587.
- PEREIRA, A.R.; ANGELOCCI, L.R.; SENTELHAS, P.C. **Agrometeorologia: fundamentos e aplicações práticas**. Guaíba: Livraria e Editora Agropecuária, 2002. 478 p.
- PETRI, J.L.; HAWERROTH, F.J.; LEITE, G.B. Fenologia de espécies silvestres de macieira como polinizadoras das cultivares Gala e Fuji. **Revista Brasileira de Fruticultura**, Jaboticabal, v.30, n.4, p.868-874, 2008.
- PILATTI, R.A.; DOVIS, V.L.; GARIGLIO, N.F.; BUYATTI, M.; MICHELOUD, N. Efecto de la fertilización foliar con Nitrógeno sobre la floración, el establecimiento de frutos y el rendimiento en cítricos. **Revista FAVE, Sección Ciencias Agrarias**, Esperanza, v.8, n.2, p.19-28, 2009.
- PIMENTEL, C.; BERNACCHI, C.; LONG, S. Limitations to photosynthesis at different temperatures in the leaves of *Citrus limon*. **Brazilian Journal of Plant Physiology**, Campinas, v.19, n.2, p.141-147, 2007.

- RIBEIRO, R.V.; CARUSO MACHADO, E.; BRUNINI, O. Ocorrência de condições ambientais para a indução do florescimento de laranjeiras no estado de São Paulo. **Revista Brasileira de Fruticultura**, Jaboticabal, v.28, n.2, p.247-253, 2006.
- RIBEIRO, R.V.; MACHADO, E.C. Some aspects of *Citrus* ecophysiology in subtropical climates: re-visiting photosynthesis under natural conditions. Review. **Brazilian Journal of Plant Physiology**, Campinas, v.19, n.4, p.393-411, 2007.
- RIBEIRO, R.V.; MACHADO, E.C.; SANTOS, M.G.; OLIVEIRA, R.F. Seasonal and diurnal changes in photosynthetic limitation of young sweet orange trees. **Environmental and Experimental Botany**, Amsterdam, v.66, p.203-211, 2009.
- RIVAS, F.; MARTÍNEZ-FUENTES, A.; MESEJO, C.; REIG, C.; AGUSTÍ, M. Efecto hormonal y nutricional del anillado en frutos de diferentes tipos de brotes de cítricos. **Agrociencia**, Montevideo, v.14, n.1, p.8-14, 2010.
- ROOSE, M.L.; GMITTER, F.G.; LEE, R.F.; HUMMER, K.E. Conservation of *Citrus* germplasm: an international survey. **Acta Horticulturae**, The Hague, v.1, n.1001, p.33-38, 2015.
- SANTOS, C.M.A.; RIBEIRO, R.V.; MAGALHÃES FILHO, J.R.; MACHADO, D.F.S.P.; MACHADO, E.C. Low substrate temperature imposes higher limitation to photosynthesis of orange plants as compared to atmospheric chilling. **Photosynthetica**, Prague, v.49, n.4, p.546-554, 2011.
- SIMÕES, D.; CABRAL, A.C.; OLIVEIRA, P.A. Citriculture economic and financial evaluation under conditions of uncertainty. **Revista Brasileira de Fruticultura**, Jaboticabal, v.37, n.4, p.859-869, 2015.
- SOUTHWICK, S.; DAVENPORT, T. Characterization of water stress and low temperature effects on flower induction in *Citrus*. **Plant Physiology**, Rockville, v.81, p.26-29, 1986.
- SYVERTSEN, J.P.; LLOYD, J.J. *Citrus*. In: SHAFFER, B.; ANDERSEN A. **Handbook of environmental physiology of fruit crops**. Boca Raton: CRC Press, 1994. v.2, p.65-99.
- TRAVADELO, M.; SORDO, M.H.; FAVARO, J.C.; PERNUZZI, C.; PERREN, R.; GARIGLIO, N.F.; MAINA, M.; ROSSLER, N.; BRIZI, M.C.; CARBONI, A. Diversificación con frambuesa: el impacto de su introducción en sistemas hortícolas de Coronda, Santa Fe, Argentina: análisis de un caso de estudio. **Revista FCA UNCUIYO**, Mendoza, v.44, n.2, p.255-262, 2012.
- WEBER, M.; CASTRO, D.; MICHELOUD, N.; BOUZO, C.; BUYATTI, M.; GARIGLIO, N. Changes in the Reproductive Traits of Low-chill Peach Tree in Response to Reproductive Shoot Pruning after Harvesting. **European Journal of Horticultural Science**, Stuttgart, v.78, n.1, p.1-7, 2013.
- YU, M.H.; DING, G.D.; GAO, G.L.; SUN, B.P.; ZHAO, Y.Y.; WAN, L.; WANG, D.Y.; GUI, Z.Y. How the plant temperature links to the air temperature in the desert plant *Artemisia ordosica*. **PLoS One**, San Francisco, v.10, n.8, 2015. Disponível em: <<http://dx.doi.org/10.1371/journal.pone.0135452>>.