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Phenological changes in olive (*Ola europaea* L.) reproductive cycle in southern Spain due to climate change

Herminia García-Mozo[1], Jose Oteros[1], Carmen Galán[1]

[1] *Department of Botany, Ecology and Plant Physiology, Agrifood Campus of International Excellence (ceiA3), University of Córdoba, Córdoba, Spain*

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Abstract

Introduction. Modifications of crop species phenology due to a changing environment are of interest because of their impact on fruit set and final harvest. Pre-flowering and flowering phenophases in olive groves at different sites of southern Spain were examined, in order to chart potential trends and determine major correlations with weather-related parameters, especially temperature and water availability. The high prevalence of olive pollen allergy in the Mediterranean population makes this study highly relevant.

Materials and methods. Ten sites in Cordoba province (Spain) during a 17-year period (1996–2012). BBCH phenology scale. Meteorological data from 1960 were analyzed; data from 1996 included on modeling analysis. Linear Mixed Models (LMMs) were developed, combining phenological and meteorological data.

Results. Since 1960, local spring temperatures have increased 1.5 °C, the number of spring rainfall days has fallen 11 days, total rainfall has declined 150 mm. Despite phenological differences between sites, attributable to altitude, phenological development during the season followed a similar pattern. Flowering dates advanced 2 days, while inflorescence emergence was delayed 24 days. Trend slopes revealed differences, an earlier period (1996–2002) with a sharp flowering advance of 15 days, and a later period (2003–2012) characterized by a gradual advance and a high bud emergence delay of 22 days.

Conclusions. LMMs was revealed as an appropriate technique for phenology behaviour analysis displaying both fixed and random interactions. Cultivars grown in the study province are adapted to climate with a synchronized response, although climate change is affecting theolive reproductive cycle in southern Spain; therefore, the timing of pollen release, with subsequent consequences on allergic population as phenological changes, could have impacts on flowering period and pollen production. Further investigation is required of the implications for crop production in Mediterranean ecosystems.

Key words

Climate change, phenology, climate, reproductive cycle, flowering, pollen, pollinosis, bud break, fruiting, *Olea europaea*

INTRODUCTION

Climate change is no longer merely projected to occur in the indeterminate future. It has already begun to be manifested in the weather regimes affecting agro-ecosystems in many regions worldwide [1]. One of the worst affected regions is the Mediterranean area, where temperatures are rising and rainfall declining; extreme rainfall events are also becoming more frequent, with longer dry periods and bursts of intense rainfall [2]. Climate affects all the physiological processes governing plant life. Plant hormones react to variations in climate, leading to changes in both vegetative and reproductive phenological phases, thus regulating the transition between the various developmental stages [3]. Those seasonal plant activities that respond both to warm and chilling conditions are likely to be modified by warmer conditions, both now and in the future [4]. The heat and chilling requirements for flowering in the olive (*Oleaeuropaea* L.) are well-documented [5, 6, 7, 8, 9]. The olive releases large amounts of pollen into the atmosphere that cause pollinosis in a high percentage of the European population, mainly located in the Mediterranean area [10, 11]. Research into airborne olive-pollen counts has provided valuable information on olive flowering behaviour and its response to a range of meteorological variables [12, 13, 14, 15, 16]. However, airborne pollen data provides information regarding only part of the reproductive cycle. Although a number of studies have addressed the influence of climate change on olive field phenology, its actual impact on allergic population remains largely unexplored as there is uncertainty about the possible impacts on flowering time and the potentiality for pollen production [14]. Moreover, most European phenological research focussing on plant responses to climate change has not included Mediterranean plants such as the olive [17, 18].

Address for correspondence: Herminia García-Mozo, Department of Botany, Ecology and Plant Physiology, Agrifood Campus of International Excellence (ceiA3), University of Córdoba, 14071 Córdoba, Spain.
E-mail: bv2gamoh@uco.es

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OBJECTIVE

The main objective of the study is to present the examination results of the response of olive reproductive phenology to climate variations. The high prevalence of olive pollinosis in the population of Spain and other Mediterranean countries makes this study especially important to understand the possible future changes on the symptoms and timing of olive pollen allergy. Particular attention is paid in the geographical changes analysing results from 10 different sites in the south of Córdoba province in southern Spain by analysing field phenological data from bud emergence to early fruiting phases, with a view to charting the influence of recent climate change and determining possible trends.

MATERIALS AND METHOD

Study area characteristics and climate. The study was carried out in the province of Córdoba, in the region of Andalusia (southern Spain). Field phenological data were collected from 1996 2012 at 3 sites located in the south of the province: Santa Cruz, Castro del Río and Baena; from 2003, 7 new sampling sites were included in the study (Fig. 1, Tab. 1). The main cultivar grown at the study sites is 'Hojiblanca', although 'Picudo' is also grown at Santa Cruz, Castro del Río and Baena (Tab. 1). This area belongs to the Mediterranean region, and local vegetation and crops are adapted to drought periods that last between 2–9 months of the year. In Córdoba city, the annual mean temperature is 17.8 °C and annual average rainfall 621 mm. Annual rainfall and mean temperature distribution for the whole of Córdoba province are shown in Figure 1. However, weather conditions vary greatly year-on-year. Data for the main meteorological variables, i.e. temperature (mean, maximum and minimum), rainfall and number of rainy days, were obtained from weather stations located near the study sites, managed by the Spanish Meteorological Agency (AEMET) '*M-a*', by the Andalusian Agroclimatic Information Network (RIA) '*M-b'*, and by the Andalusian Phytosanitary Information Alert Network (RAIF) '*M-c*', '*M-d'* and '*M-e'* (Fig. 1). For the statistical analysis of long-term trends, each study site was associated with the nearest weather station: Córdoba City and Santa Cruz were associated with *M-a;* Castro del Río with *M-c* and Baena with *M-d*.

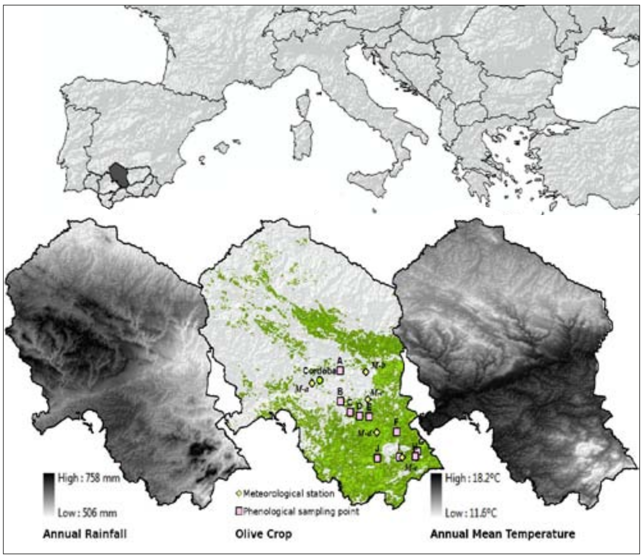

Figure 1. Location of Córdoba province. a) Annual rainfall distribution in Córdoba province. b) Olive crop distribution, location of phenological sampling sites (A-J) and weather stations (*Ma-Me*). c) Annual mean temperature distribution. Sampling-site codes: Alcolea (A), Califato (B), Santa Cruz (C), Espejo (D), Castro del Río (E), Baena (F), Fuente Tójar (G), Priego (H), Carcabuey (I) and Cabra (J)

Table 1. Site characteristics: name, coordinates, altitude, study years and observed cultivar

Site	Cultivar	Years	Altitude (m.a.s.l.)	Coordinates
Califato	Hojiblanca	2003–2012	157	37º45′N, 4º40′W
Alcolea	Hojiblanca	2002–2012	160	37º55′N, 4º40′W
Santa Cruz	Hojiblanca	1996–2012	165	37º42′N, 4º36′W
Castro	Hojiblanca	1996–2012	261	37º40′N, 4º28′W
Espejo	Hojiblanca	2003–2012	360	37º40′N, 4º32′W
Baena	Hojiblanca	1996–2012	450	37º35′N, 4º18′W
Cabra	Picudo	2003–2012	482	37º27′N, 4º25′W
Priego	Hojiblanca	2003–2012	535	37º28′N, 4º11′W
Carcabuey	Picudo	2003–2012	563	37º28′N, 4º17′W
Fuente Tójar	Picudo	2003–2012	567	37º29′N, 4º10′W

Phenology data. Reproductive phenological data for 10 olive trees at each site were analysed. Data were collected weekly from dormancy to the start of fruit development. Field observations were recorded using the international standardised BBCH scale for olive [19, 20]. The onset of each phenophase was designated by the first Julian day (day of the year) when that phenophase appears, also phenological amplitude was determined for all phenophases. The following eight BBCH phenophases were analysed:

51: Inflorescence buds starting to swell on stem.
52: Inflorescence buds open.
54: Flower clusters growing.
57: The corolla, green-coloured, is longer than the calyx.
61: Beginning of flowering: 10% of flowers open.
65: Full flowering: at least 50% of flowers open.
67: First petals falling.
68: Majority of petals fallen or faded.

Statistical analysis. Phenological behaviour during the annual reproductive cycle was examined for potential inter-site differences using cluster analysis and Linear Mixed Model (LMM) analysis for 2003–2012.

The optimum number of natural site groups by altitude was determined by hierarchical cluster analysis using Ward's method, in which information is quantified as the sum of squared distances of each element with respect to the centroid of the cluster to which it belongs. Cluster analysis was performed using the Unscrambler 9.7 software package.

To establish whether differences in phenological data might be attributable to site-group characteristics, several analyses were performed using linear mixed models (LMMs). The effect of altitude on phenology was studied as a fixed factor, grouping sites by altitude; the effect of time (years) was included as a random factor. LMMs were constructed using the InfoStat v.2012p software package.

1) To chart the inter-group differences in phenological response at a given date, a LMM model was constructed for the onset of each phenophase: the onset date was taken as the dependent variable, the effect of altitude group as fixed factor, and years as random factor.
2) Phenological amplitudes were then modelled in order to examine significant differences in seasonal phenological development due to altitude. Again, altitude groups were used as the fixed factor and years as the random factor.
3) In order to determine potential time-related differences in phenological behaviour due to altitude, the slopes of the long-term trend (2003–2012) in the 2 groups were compared. The LMM focus was also used, although – since in this case there were no random factors – analysis can be interpreted as a variance analysis between groups.

Linear trend analyses and their significance, as established by regression analysis and the F-test, were calculated for both phenological and meteorological data. For phenological data, the number of days' advance or delay for each phenophase was calculated from linear regression results, taking into account that the slope represented the average advance or delay for one year. The long-term climate trend for the city of Córdoba over the period 1960–2012 was analysed. Trend analysis was also performed for the specific weather conditions associated with study sites for 1996–2012, 1996–2002 and 2003–2012. Trends for each of the 10 study sites over the period 2003–2013 were examined. For Santa Cruz, Castro del Río and Baena, trend analysis was additionally performed for 1996–2012 and 1996–2002. The SPSS 8.0 software package was used for this purpose.

RESULTS

Climate analysis. Phenological behaviour was examined with a view to determining potential trends and their correlation with major weather-related parameters, especially temperature and water availability as expressed by rainfall. Analysis focussed on the first 6 months of each study year, since weather conditions in this period have the strongest influence on olive reproductive phenology. Analysis of climatic variations for 1960–2012 showed that, since 1960, local spring temperatures have increased by around 1.5 °C; the number of days of rainfall in the first half of the year (early spring and spring) has declined by 11 days over the same period, and recorded rainfall has dropped by around 150 mm (Tab. 2a).

During the last 17 years (1996–2012), the increase in temperature was less marked in the first 3 months (Tab. 2), and there was even a drop in January minimum temperature and March mean temperature at Santa Cruz, Castro del Río and Baena. The sharpest increase in temperature was recorded in late spring (i.e. between 1 April – 30 June), although trends at some sites (e.g. Santa Cruz) were not significant.

The study periods as a whole can be divided into 2 distinct periods as a function of climate behaviour (Tab. 2): between 1996–2002, a small increase in maximum temperature was observed in the first half of the year, although the trend was not significant, while minimum temperatures tended to fall at the lowest-altitude sites (Santa Cruz and Castro del Río). A marked decrease in rainfall was recorded throughout the province during early spring, while both total rainfall and days of rain declined considerably across the province during late spring. The most remarkable variations, however, were observed in January and May. During 2003–2012, both the amount of rainfall and number of days of rain declined during early spring, rising slightly in late spring, while March temperatures tended to decrease and April/May temperatures tended to rise.

Phenological response. The average phenological response (2003–2012) at all study sites throughout the growing season is shown in Figure 2. Phenological behaviour followed a sigmoidal curve, indicating parallel behaviour patterns at each site, with a time-lag attributable mainly to altitude. The sigmoidal shape reflects a slower response for bud phenological amplitudes (51–61), compared with the short duration of flowering phases (61–68), which pointed to a faster phenological response. Cluster analysis indicated that sites could be divided into 2 groups as a function of their topographical characteristics (Fig. 3). Linear mixed models highlighted significant differences between these 2 groups

Table 2. Slopes of the linear regression trend analysis that represent the yearly advance (-) or delay of climatic parameters
TMn – Mean Temperature (°C); TMax – Maximum Temperature (°C); TMin – Minimum Temperature (°C); Rf – Rainfall (mm); DRf – Days of Rainfall (Days)) of first 6 months of the year and also from early spring and spring periods: January – March (Jn-M), January – June (Jn-J) and April – June (A-Jn)
Table 2a. Long-term trend analysis performed in Córdoba City 1960–2012
Table 2b. Long-term trend analysis performed in Santa Cruz 1996–2012
Table 2c. Long-term trend analysis performed in Castro del Río 1996–2012
Table 2d. Long-term trend analysis performed in Baena 1996–2012

a) Córdoba		Ja	F	M	A	My	Jn	Ja-M	A-Jn	Ja-Jn
	Tmax (°C)	0.01	0.02	**0.04***	0.02	-0.00	**0.04***	**0.03***	0.02	**0.02***
	Tmin (°C)	-0.01	-0.01	**0.03***	**0.03****	**0.04****	**0.05****	0.01	**0.04****	**0.02****
1960-2012	Tmn (°C)	0.00	0.01	**0.04****	**0.03***	0.02	**0.05****	**0.02***	**0.03****	**0.02****
	Rf (mm)	-0.98	**-1.25***	-0.70	-0.23	0.35	**-0.43***	**-2.68***	-0.27	**-2.95***
	DRf (Days)	-0.12	**-0.20****	**-0.13****	-0.03	-0.01	**-0.10****	**-0.26***	-0.10	-0.22
b) Santa Cruz		Ja	F	M	A	My	Jn	Ja-M	A-Jn	Ja-Jn
	Tmax (°C)	0.03	-1.00	-0.14	-0.01	0.17	0.04	-0.07	0.07	-0.00
	Tmin (°C)	-0.14	-0.07	-0.06	0.02	0.04	0.08	-0.09	0.04	-0.02
1996–2012	Tmn (°C)	-0.05	-0.08	**-0.10***	0.00	0.10	0.06	-0.08	0.05	-0.01
	Rf (mm)	-6.28	4.11	-0.07	1.56	-1.66	-0.94	-2.25	-1.05	-3.30
	DRf (Days)	-0.31	0.32	0.06	0.01	-0.14	-0.00	0.06	-0.15	-0.06
	Tmax (°C)	0.12	0.29	-0.32	-0.30	0.29	0.32	0.03	0.10	0.07
	Tmin (°C)	-0.53	-0.27	0.22	-0.30	-0.20	0.00	-0.19	-0.17	-0.18
1996–2002	Tmn (°C)	-0.21	0.01	-0.05	-0.30	0.05	0.16	-0.08	-0.03	-0.06
	Rf (mm)	-28.45	-4.50	**25.09***	6.04	**-15.46***	-5.44	-7.86	-14.85	-22.74
	DRf (Days)	-1.11	0.18	1.18	-0.11	-0.46	-0.36	0.24	-0.93	-0.66
	Tmax (°C)	-0.00	0.11	0.06	0.04	0.08	-0.18	0.06	-0.02	0.02
	Tmin (°C)	0.18	-0.15	-1.00	0.09	0.09	-0.09	-0.02	0.03	0.01
2003–2012	Tmn (°C)	0.09	-0.02	-0.02	0.06	0.09	-0.13	0.02	0.01	0.01
	Rf (mm)	1.96	-1.18	-2.03	3.39	0.80	1.47	-1.26	5.67	4.44
	DRf (Days)	0.19	-0.33	-0.27	0.33	0.12	0.20	-0.42	0.66	0.24

Table 2. Slopes of the linear regression trend analysis that represent the yearly advance (-) or delay of climatic parameters (Continuation)

c) Castro del Río		Ja	F	M	A	My	Jn	Ja-M	A-Jn	Ja-Jn
1996–2012	Tmax (°C)	0.08	-0.10	-0.16	-0.02	0.11	-0.07	-0.06	0.01	-0.03
	Tmin (°C)	-0.22	-0.18	**-0.24***	-0.12	-0.09	-0.08	**-0.21***	-0.10	**-0.16***
	Tmn (°C)	-0.07	-0.14	**-0.20****	-0.07	0.01	-0.07	-0.14	-0.05	-0.09
	Rf (mm)	-4.38	2.35	0.47	2.72	-0.42	-0.16	-1.56	2.13	0.60
	DRf (Days)	-0.47	0.28	0.13	0.08	-0.08	0.04	-0.06	0.03	-0.06
1996–2002	Tmax (°C)	0.24	0.37	-0.30	-0.25	0.46	0.23	0.11	0.15	0.13
	Tmin (°C)	-0.31	-0.17	0.36	-0.07	0.01	-0.06	-0.04	-0.04	-0.04
	Tmn (°C)	-0.03	0.10	0.03	-0.16	0.23	0.09	0.03	0.05	0.04
	Rf (mm)	-14.63	-2.95	**14.61***	5.86	-12.53	-2.84	-2.97	-9.51	-12.48
	DRf (Days)	-1.46	-0.29	0.75	-0.07	-0.14	0.00	-0.99	-0.21	-1.20
2003–2012	Tmax (°C)	0.33	0.18	0.08	0.06	0.09	-0.28	0.19	-0.05	0.07
	Tmin (°C)	0.36	-0.04	-0.11	0.14	0.08	-0.09	0.07	0.04	0.06
	Tmn (°C)	0.34	0.07	-0.02	0.10	0.08	-0.19	0.13	-0.00	0.07
	Rf (mm)	-0.04	-5.30	-2.71	5.33	3.83	0.98	-8.04	10.14	2.10
	DRf (Days)	-0.33	-0.32	-0.02	0.17	0.08	0.09	-0.66	0.36	-0.30
d) Baena		Ja	F	M	A	My	Jn	Ja-M	A-Jn	Ja-Jn
1996–2012	Tmax (°C)	-0.03	-0.06	-0.08	0.10	**0.26***	0.13	-0.06	**0.16***	0.05
	Tmin (°C)	-0.16	-0.11	**-0.10***	0.01	0.06	0.06	**-0.13***	0.04	-0.04
	Tmn (°C)	-0.10	-0.09	-0.09	0.05	**0.16***	0.10	-0.09	**0.10***	0.00
	Rf (mm)	-10.08	2.27	-0.11	2.42	-2.61	1.18	-7.92	0.99	-6.90
	DRf (Days)	-0.38	0.15	0.14	0.07	-0.15	0.02	-0.09	-0.06	-0.18
1996–2002	Tmax (°C)	0.32	0.26	-0.23	-0.27	0.27	0.40	0.12	0.13	0.12
	Tmin (°C)	0.02	0.13	0.22	-0.17	0.02	0.17	0.12	0.01	0.07
	Tmn (°C)	0.17	0.19	-0.01	-0.22	0.15	0.29	0.12	0.07	0.09
	Rf (mm)	-46.86	-3.48	20.14	5.90	-13.50	-2.53	-30.21	-10.14	-40.32
	DRf (Days)	-0.89	0.29	0.96	-0.04	-0.18	-0.29	0.36	-0.51	-0.12
2003–2012	Tmax (°C)	0.00	0.05	0.03	0.07	0.12	-0.19	0.03	0.00	0.02
	Tmin (°C)	0.15	-0.11	-0.09	0.13	0.06	-0.04	-0.02	0.05	0.02
	Tmn (°C)	0.08	-0.03	-0.03	0.10	0.09	-0.11	0.01	0.03	0.02
	Rf (mm)	3.85	-0.13	-5.83	5.91	0.10	3.73	-2.10	9.75	7.62
	DRf (Days)	0.19	-0.25	-0.18	0.21	-0.04	0.09	-0.24	0.27	0.03

*p<0.05;** p<0.01

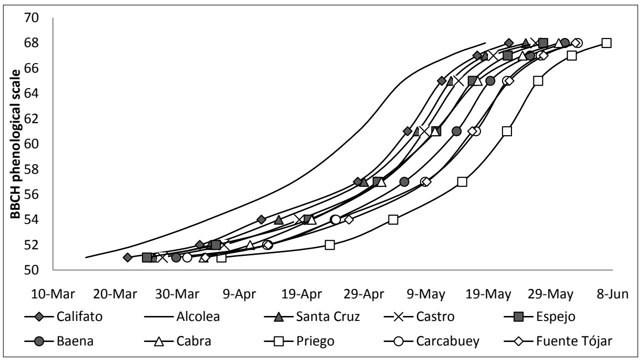

Figure 2. Average phenological development throughout the growing season at the different study sites

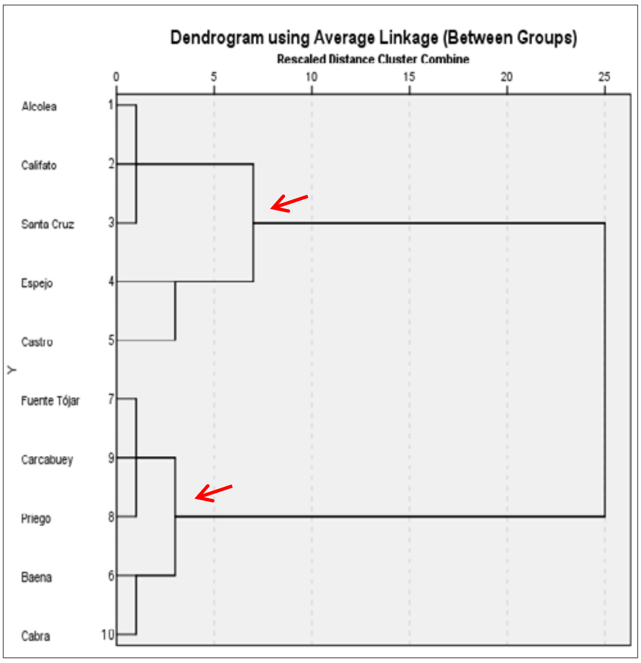

Figure 3. Cluster analysis of phenological sampling sites grouped by altitude, showing two groups

for the onset of every phenophase (Tab. 3). Comparison of phenological amplitudes between the 2 groups revealed no significant differences, suggesting that phenological behaviour (i.e. the period elapsing between phenophases) in the olive was similar throughout the province (Tab. 3). Linear mixed models also pointed to a similar long-term trend, although altitude-related differences in trend were found for 2 phenophases: inflorescence buds open (52) and full flowering (65).

Table 3. Linear Mixed Models summary. Mixed Models compare phenological features of 2 groups of sampling points grouped by altitude; in this sense, phenological differences due to altitude are shown. Differences between groups in the main phenophases (*Pheno*) were tested, as well as differences in phenological ranges (*Range*) and differences in slopes of long-term trends shown for each phenophase (*Trend*). N – Number of cases; nDf – degrees of freedom; nDf – numerator; dDf – denominator degrees of freedom; AIC – Akaike information criterion scores; p – P-values of 'group fixed factor' on every model. Significant differences between groups are indicated in bold letter (p<0.01)

Linear Mixed Models Summary															
Pheno	*N*	*nDF*	*dDF*	*AIC*	*P*	*Range*	*N*	*nDF*	*dDF*	*AIC*	*p*	*Trend*	*N*	*AIC*	*p*
51	100	1	89	703.56	**<0.0001**	51–52	100	1	89	605.93	0.0351	51	10	23.06	**0.0012**
52	100	1	89	713.48	**<0.0001**	52–54	100	1	89	557.21	0.2006	52	10	30.13	0.0905
54	100	1	89	708.33	**<0.0001**	54–57	100	1	89	621.18	0.1624	54	10	26.66	0.0617
57	100	1	89	697.80	**<0.0001**	57–61	100	1	89	606.41	0.0265	57	10	27.86	0.887
61	100	1	89	643.91	**<0.0001**	61–65	100	1	89	451.24	0.6044	61	10	16.38	0.2457
65	100	1	89	625.73	**<0.0001**	65–67	100	1	89	434.85	0.9589	65	10	12.96	**0.0083**
67	100	1	89	612.33	**<0.0001**	67–68	100	1	89	436.68	0.2794	67	10	16.22	0.0257
68	100	1	89	621.18	**<0.0001**	51–61	100	1	89	682.06	0.6796	68	10	18.85	0.1862

Phenological trends. Over the last 17 years, the start of flowering (61), flowering (65) and fruiting (68) have occurred gradually earlier, displaying an average advance of 1.7 days, 1.87 days and 3.4 days, respectively. By contrast, a marked delay of 24 days has been recorded in bud emergence (51; Table 4). Pre-flowering amplitude (51–61) and flowering amplitude (61–67) have been shortened by 26 days and 38 days, respectively.

As indicated earlier, the only altitude-related difference in phenological trends was a delay recorded for phenophases 52 and 65 at the highest altitudes. Nevertheless, different trends were noted for certain phenophases, for example, flowering start (61) and full flowering (65) advanced at different speeds, and also between periods: all phenophases advanced in 1996–2002, but not in 2003–2012 (Fig. 4).

The delay in the onset of phenophase 51 during 1996–2012 was more marked in the latter years (2003–2012) than in the early years (1996–2003). Between 1996–2002, there was a notable advance in flowering (61 and 65), whereas between 2003–2012 the advance was more gradual. Fruit emergence (68) occurred 12.6 days earlier in 1996–2002 and 2.3 days earlier in 2003–2012. Phenological amplitudes displayed differing behaviour in the 2 periods, lengthening in 1996–2002 and shortening in 2003–2012 (Tab. 4).

Analysis of phenological trends during 1996–2012 indicated an advance in flowering phenophases, but not in the phenological range from bud break to just before anthesis, the duration of which actually increased. The increase in spring temperatures, especially in April,

Table 4. Slopes of linear regression trend analysis that represent the yearly advance (-) or delay of phenological phases and yearly shortening (-) or elongation of phenological ranges

Study years	Localty	Phenophase							
		51	52	61	65	68	51–61	61–67	51–68
1996–2012	Santa Cruz	**1.74****	0.54	0.21	0.09	-0.17	**-1.36***	**-2.34***	-0.47
	Castro del Río	**1.44***	0.04	-0.33	-0.23	-0.08	**-1.64***	**-2.13***	0.28
	Baena	1.06	-0.24	-0.19	-0.18	-0.36	**-1.62***	**-2.35***	0.04
	Average	***1.41****	*0.12*	*-0.10*	*-0.11*	*-0.20*	***-1.54****	***-2.29****	*-0.05*
1996–2002	Santa Cruz	-3.90	**-4.25***	-2.32	-2.61	-2.26	0.20	4.50	**1.16****
	Castro del Río	-4.90	-2.68	-2.61	-2.18	-1.58	0.80	**9.10***	**2.37***
	Baena	-7.90	-5.32	-1.93	-1.61	-1.59	0.60	9.40	3.04
	Average	*-5.57*	***-4.08****	*-2.28*	*-2.13*	*-1.80*	*0.54*	*7.60*	***2.19****
2003–2012	Santa Cruz	**1.93***	2.13	0.04	-0.23	-0.35	**-1.87***	-0.31	**-3.04***
	Castro del Río	1.59	0.66	-0.81	-0.85	-0.30	**-2.34***	0.28	**-2.59***
	Baena	**3.18***	1.72	0.13	0.24	-0.04	**-2.99***	-0.04	**-3.24****
	Average	***2.22****	*1.52*	*-0.21*	*-0.28*	*-0.23*	***-2.39****	*-0.02*	***-2.95*****
	Califato	0.58	0.25	-0.30	-0.55	-0.67	-1.32	-0.50	**-2.35***
	Alcolea	1.08	1.44	0.12	-0.68	-1.27	-0.85	**-1.16***	**-3.21***
	Espejo	1.96	1.41	0.00	-0.24	0.16	**-2.43***	0.11	**-2.93***
	Cabra	**3.05***	**3.92***	0.42	0.12	-0.13	**-2.95****	-0.42	**-4.40****
	Priego	**3.72****	1.17	0.50	0.67	0.33	**-3.20****	-0.06	**-3.49****
	Carcabuey	**3.71****	2.25	-0.08	0.04	0.02	**-3.68***	0.35	**-3.67****
	Fuente Tójar	**2.37****	2.27	-0.42	-0.25	-0.59	**-2.61***	0.01	**-4.07****
	All sites average	***2.31*****	*1.75*	*-0.03*	*-0.23*	*-0.29*	***-2.42*****	*-0.16*	***-3.28*****

*p<0.05;** p<0.01

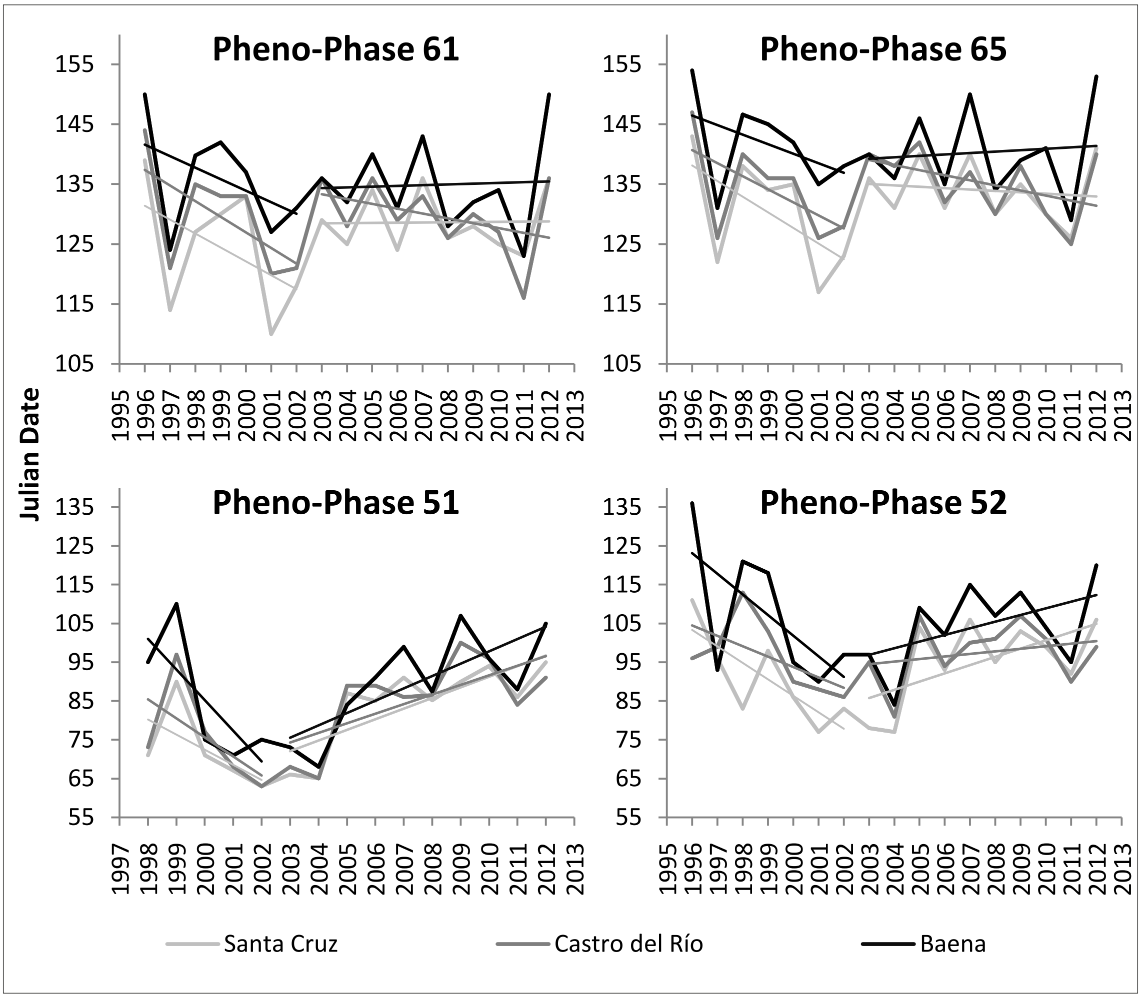

Figure 4. Temporal trends for various phenophases, 51: Inflorescence buds start to swell on stem; 52: Inflorescence buds open; 61: Start of flowering: 10% of flowers open; 65: Full flowering: at least 50% of flowers open. Straight lines represent linear regression trends. Different phenological trends were found for the periods 1996–2002 and 2003–2012

coincided with a decline in the flowering slope, while the increase in minimum temperature coincided with a delay in the onset of the bud break period. Finally, the correlation between temperature and the advance of flowering and the lengthening of the pre-flowering period was more marked from 1996–2002 than from 2003–2012.

DISCUSSION

LMMs are the most appropriate technique for studying phenomena displaying both fixed and random interactions. Here, the technique was applied to field phenological data obtained during the main olive reproductive cycle and covering bud break, flowering and early fruiting. The large number of records available for this multi-year study enabled a detailed analysis of the olive's response to recent climate change in southern Spain. The phenological response to temperature variations in spring, and especially in early spring, is unquestionable. Although linear mixed models revealed significant phenological differences between sites at a given date, mostly attributable to altitude, phenological development patterns over the course of the season were similar through the province. In this sense, altitude impacts on the airborne pollen exposure period of local population. No significant differences were observed between the slope of curves, or in the duration of the phenological range; behaviour patterns were parallel, except for a time-lag at higher altitudes reflecting the influence of lower temperatures at those sites. Spatial variations in tree phenology are influenced by genetic variations among individuals. The local climate has prompted a degree of natural selection, leading to specific adaptations. Cultivars grown in the study province appear to be perfectly adapted to the climate, displaying a synchronized response. At sites where the 'Picudo' cultivar is grown, the phenological response took place earlier than that of 'Hojiblanca' trees growing at the same altitude, probably reflecting the greater temperature sensitivity of this cultivar, which is well adapted to colder areas. Previous research into the phenological response of different cultivars suggests that the 'Picudo' cultivar in Córdoba province is more affected by temperature and less controlled by photoperiod than other cultivars. The characteristic sigmoidal shape of the reproductive phenology curve may be due to the fact that during the early bud phases the response to climate is slower than during the flowering phases, where a faster phenological response is recorded.

Flowering is occurring progressively earlier which could anticipate and increase health crisis by allergenic pollen, a finding which supports predictions made by a number of authors some years ago [13, 21]. By contrast, an observed delay in bud break date may be related to the recorded increase in minimum temperatures, which prolongs the time required for chilling-unit accumulation prior to bud emergence [22]. These variations on pre-flowering conditions could have implications on the pollen production with unknown implications on allergenic population. Indeed, the rise in minimum temperatures was particularly significant during the latter part of the study period, coinciding with a more marked delay in the onset of bud emergence. The advance in flowering and fruiting is wholly in accordance with phenological data reported across Europe indicating a mean advance of 2.5 days [18, 23]. This is an important fact to be taken into account by allergists and patients in the Mediterranean area. Moreover, it is a more marked advance than that detected in tree phenology in Central Europe [24]. However, the presented results contrast with those reported by Vitasse et al. [25], who suggest that climate change will lengthen tree life cycles in the northern hemisphere due to the advance of bud burst and flowering. The present findings point primarily to a shortening of the olive reproductive cycle which may have a number of implications, among them a change in the exposure to airborne allergens, a higher risk of freezing, an impaired competitive ability to metabolize nutrients, and an increased risk of non-fertile flowers and pollen abortion. The main consequence of the decrease in rainfall, especially in May – a period widely considered crucial for pollination, fruit setting and therefore final olive production – may be a declining olive crop [26, 27, 28].

The presented findings appear to provide another indication that climate change is a major contributor to the recent changes suffered by agro-ecosystems, particularly in the Mediterranean area. These changes, in the case of anemophilous species with extremely allergenic pollen, such as olive tree, could have strong impacts on allergic population. The delay in pre-flowering phenophases could have implications on the potential pollen productivity due to changes on growing conditions. Also, the advance in flowering period could anticipate and increase health crisis by pollinosis. On the other hand, the shortening of the pollination period could decrease exposure time and reduce harmful effects.

CONCLUSIONS

Although significant phenological differences were recorded between study sites on a given date, mostly due to differences in altitude, phenological development over the season was similar throughout the province, displaying parallel behaviour with an altitude-linked time-lag. Long-term trends across the province were similar, apart from altitude-related differences in trends for the onset of 2 phenophases – 52 and 65. This means that the altitudinal features of a place impacts directly on potential pollinosis symptoms of local population, mainly on the duration and start of allergy season.

Phenological trend analysis showed that flowering advanced by an average of 2 days during the study period, while inflorescence emergence was delayed by roughly 24 days. Examination of trend slopes revealed differences between an earlier period (1996–2002) marked by a sharp advance in flowering (15 days), and a later period (2003–2012) characterised by a more gradual advance in flowering, together with a considerable delay in bud emergence (22 days). These phenological changes could cause strong impacts the on allergic population, with the advance on the allergens exposure and the shortening of the flowering period. A correlation was observed between the flowering slope and the increase in spring temperatures, especially in April, while the increase in minimum temperatures was associated with a delay in budburst.

Climate change is clearly affecting the olive reproductive cycle, with subsequent consequences on the medical impact of the pollen release timing and agronomic impact related to olive fruit production.

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