

Study of Poaceae phenology in a Mediterranean climate. Which species contribute most to airborne pollen counts?

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Abstract The present study sought to determine which of the common Poaceae species in the study area contribute most to the Poaceae pollen season curve, and to determine the phenological behaviour of the species studied. The different floral phenophases in thirty-three Poaceae species common in and around the city of Córdoba (SW Iberian Peninsula) were checked periodically over the period 2004–2006. Results showed that longer phenological ranges were recorded in the coolest and wettest year, and shorter ranges in the warmest and driest year. Moreover, ranges varied as a function of altitude: populations in lower-lying areas flowered earlier than those at higher altitudes. The results, taken in conjunction with the findings of preliminary research into potential pollen production, showed that probably only four of the Poaceae species studied—*Dactylis glomerata*, *Lolium rigidum*, *Trisetaria panicea* and *Vulpia geniculata*—were major contributors to the Poaceae airborne pollen curve.

Keywords Phenology · Poaceae ·
Pollen · Phenological phases

1 Introduction

Phenology is the study of recurrent biological phenomena, the biotic and non-biotic factors that cause them, and the interrelation between these phenomena in the same or in various species (Lieth 1974). Phenological studies are based on direct observation of the annual life-cycle of species (Schwartz 1999; Menzel 2000, 2002). The observation unit may vary from a single species to a full ecosystem, and may comprise a small or a large area. Observation of these processes requires the prior division of the whole event into specific phenophases. The occurrence of a phenophase is determined by the biotic characteristics of the species and by local climate characteristics (Orshan 1989). A strong correspondence exists between climate and phenological patterns in almost all ecosystems that exhibit seasonal climate variation (Rathcke and Lacey 1985).

The Poaceae family comprises more than 700 genera containing around 10,000 species (Watson and Dallwitz 1992). It includes both annual and perennial herbs, which are essentially anemophilous. In most cases, grasses present a high number of flowers per inflorescence that release a certain number of pollen grains into the atmosphere. Many species are well distributed in and around cities; this wide distribution, together with the elevated allergenicity of grass pollen grains, makes these species the main cause of pollinosis in Europe in general (D'Amato et al. 2007), and particularly in Spain (Jato et al. 2009).

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Airborne pollen counts at any given moment are influenced by factors related to plant pollen production, and by a number of aerobiological processes. Each of the sequential steps in the aerobiological pathway is linked to different biological events in association with atmospheric phenomena. Airborne pollen counts and timing depend on the floral phenology of the emitting species.

In 2003, Prieto Baena et al. studied pollen production per inflorescence in more than 30 different Poaceae species, to identify the main pollen producers. To complete this research, the main goal of the present study was to compare the Poaceae pollen season with the floral phenology of the same Poaceae species, in order to identify those species contributing most to the pollen curve.

2 Materials and methods

This study was carried out from 2004 to 2006. The study area is located in the mountains to the north of Córdoba (Sierra Morena), and in Córdoba city (southwestern Iberian Peninsula). The climate is Mediterranean with some continental features, and is characterized by temperate-cold winters and hot dry summers, with an annual average temperature of 17.6°C and an annual average rainfall of 535 mm for the period 1971–2000 (Agencia Estatal de Meteorología 2001).

2.1 Weather data

Weather data, supplied by the Andalusian Regional Government Agroclimatic Information Network, were obtained at a weather station on the outskirts of Córdoba city (UTM coordinates X: 341642.0; Y: 4192085.0; Altitude: 117 m.a.s.l.). Major weather parameters, i.e. rainfall, average temperature, and average humidity during the survey season, were taken into account when performing the meteorological analysis.

2.2 Aerobiology

Airborne Poaceae pollen counts were obtained using a Hirst-type sampler (Hirst 1952) located on the roof of the Education Sciences Faculty building, in the city of Córdoba. Data were obtained following the

protocol recommended by the Spanish Aerobiology Network (REA) in its Management and Quality Manual (Galán et al. 2007).

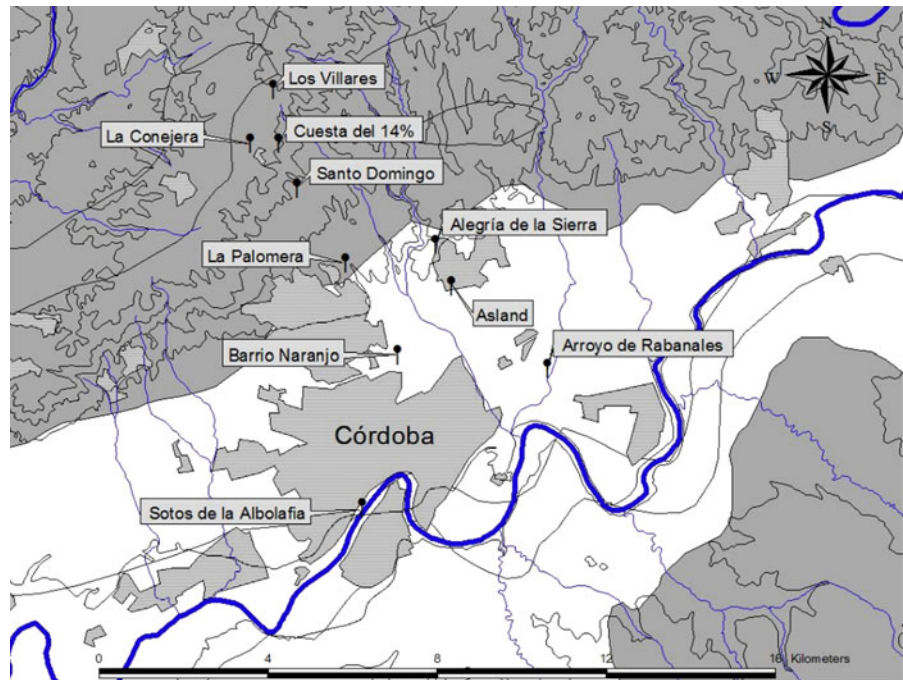
In order to compare airborne pollen counts with flowering phenology, the pollen season was taken as the period from the first day on which an average of 50 pollen grains/m³ of air was recorded to the last day when that count was recorded. This pollen concentration is considered by the Spanish Aerobiology Network as the threshold required for a large percentage of the susceptible population to develop allergy symptoms associated with the presence of this type of pollen in the air (Galán et al. 2007).

2.3 Phenological data

Phenological data were obtained from ten different sampling points, representing the main habitats where grasses grow (Fig. 1). Table 1 shows the UTM coordinates and altitude of these sampling points. From March to June, phenological observations were carried out weekly. The following phenological phases, based on the Barbieri system (Barbieri et al. 1989), were observed (Fig. 2) and translated into BBCH phenological phases to facilitate international scientific communication: Preflowering phase or Phase 0 (BBCH 60): this comprises the period of time between plant emergence and the start of flowering; Start of flowering phase or Phase 1 (approx. < BBCH 62): begins when first open florets are observed and ends when 25% of florets are open; Full flowering Phase or Phase 2 (approx. < BBCH 67): comprises the period of time in which between 25 and 75% of florets are open. End of flowering phase or Phase 3 (approx. < BBCH 69): starts when 75% of florets are open and ends when all florets have released all their pollen grains. Fructification phase or Phase 4 (BBCH 69): starts when all florets have released all their pollen grains.

When calculating the length of the various phenophases, the number of days elapsing from 1 March to the start of phase 0 was also calculated as time (V). This value reflects both differences in flowering time between species and year-on-year differences for the same species.

Thirty-three different Poaceae species were studied: *Aegilops geniculata* Roth., *Aegilops neglecta* Req. ex Bertol, *Aegilops triuncialis* L., *Agrostis pourretii* Willd., *Anthoxanthum aristatum* Boiss.,

Fig. 1 Distribution of sampling points**Table 1** Sampling points: UTM coordinates and altitude

Sampling point	UTM X	UTM Y	Altitude (m.a.s.l.)
Los Villares	341149	4203129	585
La Conejera	340584	4201852	555
Cuesta del 14%	341181	4201771	511
Santo Domingo	341758	4200722	368
La Palomera	342737	4198985	251
Alegría de la Sierra	344926	4199407	217
Asland	345252	4198511	184
Barrio Naranjo	344300	4196752	146
Arroyo de Rabanales	357483	4196492	109
Sotos de la Albolafia	343274	4193395	92

Arrhenatherum album (Vahl.) W.D. Clayton, *Avena barbata* Pott ex Link., *Avena sterilis* L., *Brachypodium dystachion* (L.) Beauv., *Briza maxima* L., *Briza minor* L., *Bromus diandrus* Roth., *Bromus hordeaceus* L., *Bromus lanceolatus* Roth., *Bromus matritensis* L., *Cynosurus echinatus* L., *Dactylis glomerata* L., *Elymus repens* (L.) Gould, *Festuca arundinacea* Schreber, *Gaudinia fragilis* (L.) Beauv., *Holcus setiglumis* Boiss & Reuter, *Hordeum geniculatum* All., *Hordeum leporinum* Link., *Hyparrhenia hirta* (L.) Stapf in Oliver, *Lolium rigidum* Gaudin, *Phalaris minor* Retz, *Poa trivialis* L., *Rostraria cristata* (L.)

Tzvelez, *Stipa capensis* Thumb, *Taeniatherium caput-medusae* (L.) Nevski, *Trisetaria panicea* (Lam.) Paunero, *Vulpia geniculata* (L.) Link, and *Vulpia myuros* (L.) C.C. Gmelin. Only those species displaying a coverage of at least 25 individuals/m² for each population were taken into account. For that reason, some species were surveyed in only one or two of the three study years.

2.4 Statistical analysis

Statistical analysis was carried out to determine the relationship between flowering phenology and airborne pollen counts. Shapiro-Wilk's W test was used to check the approximation of the frequency distribution of collected data to a normal curve. Since data could not be fitted to a Gaussian curve, phenological data and airborne pollen counts were compared by non-parametric correlation analysis (Spearman correlation test). The pollen data used for analyses were the accumulated data for the corresponding 7-day period. Phenological data were transformed to enable analysis of correlations with airborne pollen data, as follows:

- Data greater than two (full flowering phase) were subtracted from four (fructification phase, end of



Fig. 2 Phenological phases in *Trisetaria panicea*. **a** Preflowering phase or Phase 0. **b** Start of Flowering phase or Phase 1. **c** Full Flowering phase or Phase 2. **d** Phase 2, spike detail. **e**

End of Flowering phase or Phase 3. **f** Phase 3, spike detail. **g** Fructification phase or Phase 4

flowering phase) assuming that an increase in the phenological phase after full flowering involved a decline in pollen release.

- b. Phenological values were increased by one unit in order to use the zero value as an indicator of data absence.

The numerical equivalence used for the statistical analysis for each phase is shown in Table 2.

3 Results

3.1 Meteorology and aerobiology

Data for monthly accumulated rainfall, average temperature, and average relative humidity over the survey season are shown in Fig. 3. The results showed that 2004 was the coolest and wettest year

Table 2 Numerical equivalence for each phenological phase used in the statistical analysis

Phase	Numerical equivalence
Phase 0	1
Phase 1	2
Phase 2	3
Phase 3	2
Phase 4	1

Phase 0: Preflowering Phase, Phase 1: Start of flowering, Phase 2: Full flowering, Phase 3: End of flowering, Phase 4: Fructification phase

in the series, while year 2005 was the warmest and driest.

Weekly pollen counts over the survey season for each study year are shown in Fig. 4. The highest counts were recorded in 2004, and the lowest in 2005. In 2004, the highest counts were recorded during the last week in May (28.05–03.06), after a rainy period, whereas in 2005 they were recorded the last week in April. In 2006, similar counts were recorded during the first 3 weeks of May, these being the highest of the survey season.

Daily pollen counts and rainfall during the survey season are shown in Fig. 5. In general, the highest daily pollen counts were recorded from early May to early June, except in 2004 when the peak period ended in mid-June. These high counts were usually recorded after the main spring rainfall period, except in 2005, when rainfall during this period was scarce. Finally, the three well-defined peaks observed in pollen curves are probably attributable to differences in the flowering period of different species.

3.2 Phenology and aerobiology

The different phenophase ranges for each species were calculated over the 3-year study period. This value was obtained by adding together the range of each phenophase for each population and dividing the result by the number of populations of a given species. The results are shown in Table 3. In general, over the three study years some species started the flowering phase in early spring (mid- to end-March), e.g. *Bromus dianthus*, *B. hordeaceus*, *B. matritensis* and *Hordeum leporinum*. Others, however, started their flowering phase later, in late April, e.g. *Cynosurus echinatus*,

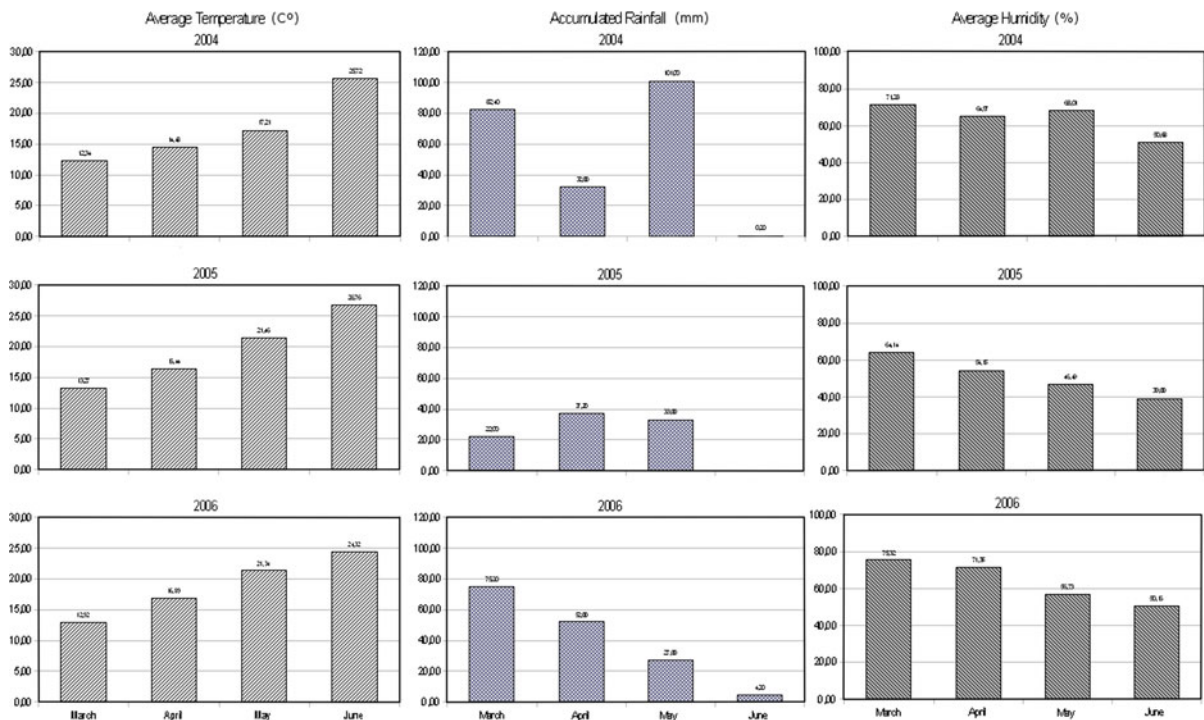
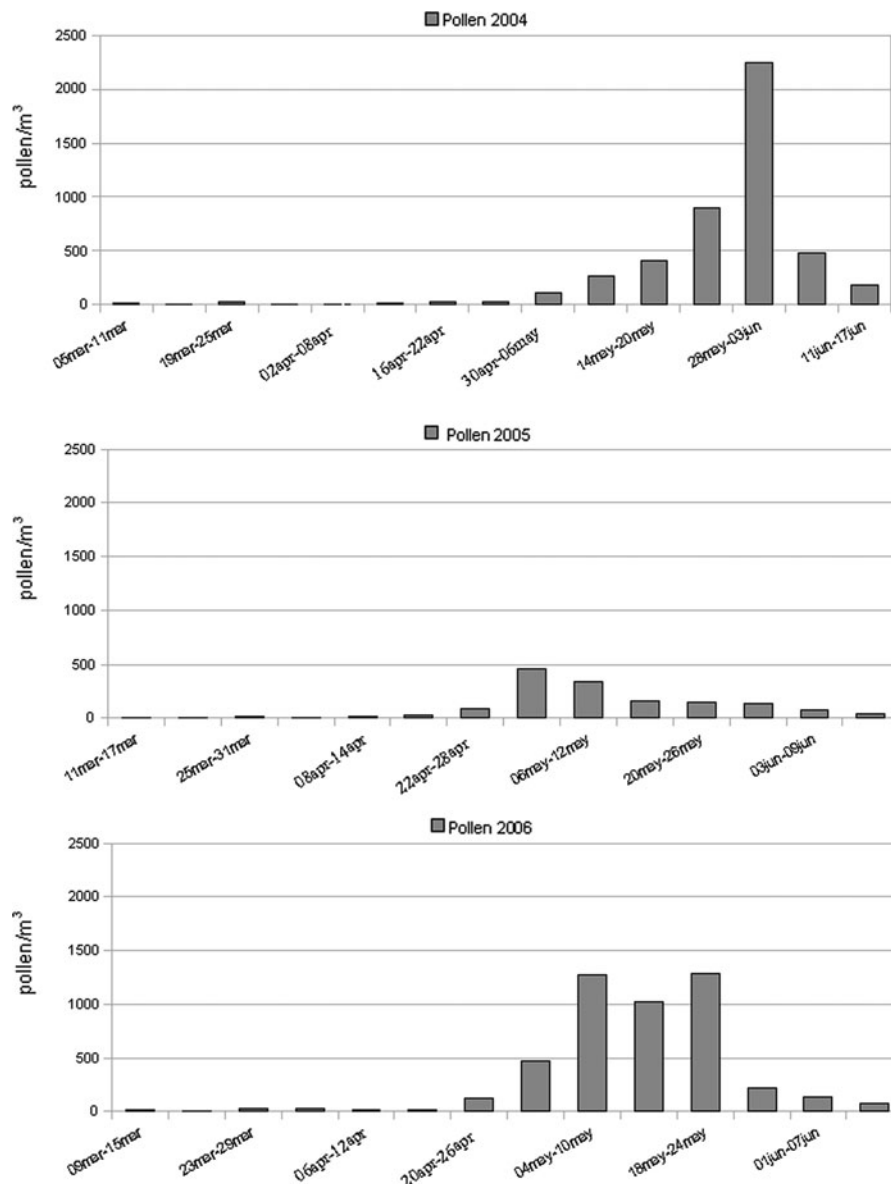


Fig. 3 Monthly average temperature, accumulated rainfall and humidity. Years 2004–2006

Fig. 4 Accumulated weekly pollen counts



Dactylis glomerata, *Gaudinia fragilis*. Some species, including *Avena barbata*, *A. sterilis*, *Bromus diandrus*, *B. matritensis*, *Hordeum leporinum* *Hyparrhenia hirta* and *Vulpia geniculata*, displayed a longer-than-average phenological range, while for others, such as *Cynosurus echinatus*, *Trisetaria panicea* and *Gaudinia fragilis*, the range was shorter. The full-flowering phase was generally the longest, and the preflowering phase the shortest.

Figures 6, 7, 8 show the length of the different phenophases for each study population (horizontal bars). Pollen-season start and end are also shown

(continuous vertical bars), while the peak day is indicated by a broken vertical bar, in order to distinguish those populations contributing most to the Poaceae pollen curve, i.e. those populations whose full-flowering phase occurred during the main pollen season and close to the peak day.

In 2004, populations at the highest altitudes generally appeared to display a closer coincidence of full flowering phase and peak airborne pollen counts than those located at lower altitudes. This was particularly true of *Brachypodium dystachion*, *Dactylis glomerata*, *Lolium rigidum*, *Stipa capensis*, *Trisetaria panicea*, and *Vulpia geniculata*.

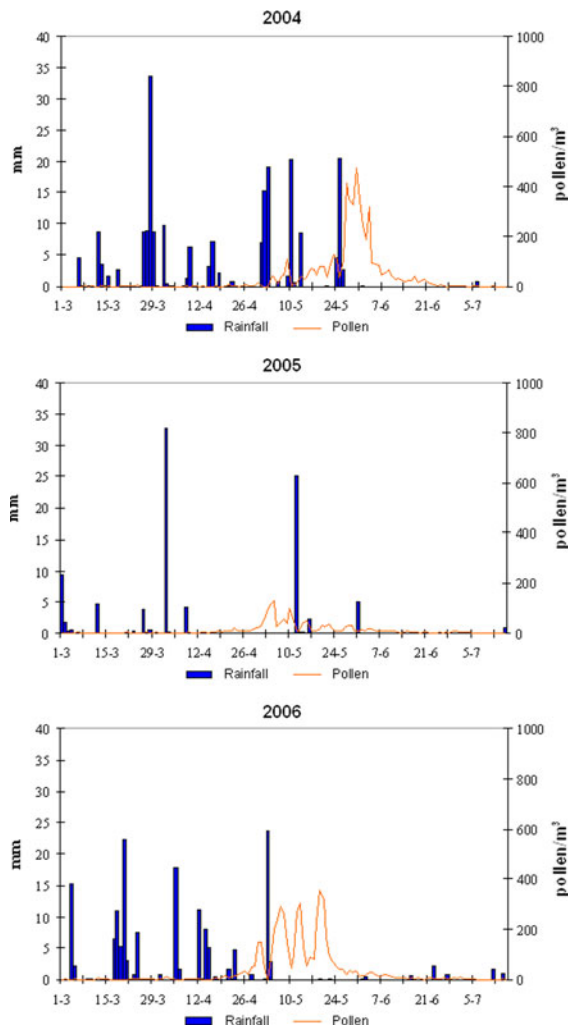


Fig. 5 Daily pollen counts and rainfall

In 2005, coincidence was greater for populations in mid-altitude areas (in the foothills of the mountains), than for those at either higher or lower altitudes; the greatest degree of coincidence between full flowering phase and peak airborne pollen counts was found for *Avena barbata*, *Bromus hordeaceus*, *Brachypodium dystachion*, *Dactylis glomerata*, *Stipa capensis*, *Taeniatherium caput-medusae*, *Trisetaria panicea*, and *Vulpia geniculata*. Finally, in 2006 the greatest coincidence was found for the highest-altitude populations, particularly in *Aegilops geniculata*, *Avena barbata*, *Bromus hordeaceus*, *Brachypodium dystachion*, *Dactylis glomerata*, *Stipa capensis*, *Taeniatherium caput-medusae*, *Trisetaria panicea*, *Vulpia geniculata* and *V. myuros*.

In summary, in all three studies years *Brachypodium dystachion*, *Dactylis glomerata*, *Lolium rigidum*, *Stipa capensis*, *Trisetaria panicea* and *Vulpia geniculata* displayed a high degree of coincidence between full flowering period and main pollen season, particularly in high altitude areas.

The second step of the phenological study focussed on analysing potential correlations between the weekly phenological indices for those species displaying the greatest coincidence between full flowering period and main pollen season, and accumulated weekly Poaceae pollen counts. The results showed strongly-significant correlations between phenological indices for most populations and weekly pollen indices in all three study years (Tables 4, 5, 6).

4 Discussion

The grass flowering season can vary from year to year depending on local weather conditions, mainly in terms of an advance or delay in phenophases (Puppi Branzi and Zanotti 1992). Environmental factors such as rainfall, temperature, and solar radiation are responsible for the major year-on-year variations (Sánchez-Mesa et al. 2005; Cenci and Ceschia 2000). Phenological observations in *Quercus* sp. show that catkin frost damage must be taken into account as a factor influencing low pollen production: during a study of floral phenology and pollen production in *Quercus suber* L. an interruption of catkin development was detected due to a sharp drop in minimum temperatures. Cold spells during flowering may be one the factors influencing the high interannual variability observed in *Quercus* pollen records (García-Mozo et al. 2001). The flowering phenology of *Quercus* species in different locations of Córdoba, Spain, showed shorter ranges in years with warmer springs and longer ranges in colder years. Phenological ranges differed as a function of species and location. Individuals located at lower altitudes and facing the sun flowered earlier (Gómez Casero et al. 2007). Here, the particular weather features in 2004 (rainier and cooler than the other 2 years) prompted an advance in the flowering phenology of Poaceae species. Most species also displayed their longest phenophase lengths in 2004, and—as a result—the longest pollen season and highest pollen counts. By contrast, in 2005 the high average temperature and

Table 3 Average length of different phenophases in each species studied

Species	Sampling points			V			Phase 0			Phase 1			Phase 2			Phase 3			Range		
	2004	2005	2006	2004	2005	2006	2004	2005	2006	2004	2005	2006	2004	2005	2006	2004	2005	2006	2004	2005	2006
<i>Aegilops geniculata</i>	4	3	4	42.3	57.7	48.7	5.7	6.7	12	14.3	7.7	10	15	6.3	14.3	11.7	7.3	8	46.7	28	44.3
<i>Arrhenatherum album</i>	2	2	2	50.5	53	42.6	14	4	9.2	14	6.5	10	13	9.5	15	8	8	12	49	28	46.2
<i>Avena barbata</i>	7	6	7	30.2	46	37	11.4	7	14	13.6	9.6	11	17.6	11.6	16	12	9.6	7	54.6	37.8	49
<i>Avena sterilis</i>	1	1	1	26	39	47.5	11	14	12.5	14	12	11	17	12	15	14	11	7	56	49	45.5
<i>Brachypodium distachion</i>	5	5	5	51.2	53	49.6	12.2	7.6	11.2	14.8	7.8	10.4	12.4	6.8	12.8	8.2	7.2	7.6	47.6	29.4	42
<i>Briza maxima</i>	2	2	2	43.5	49.5	44	6	5.5	7.5	13.5	7	14	17.5	9	14	12	6.5	10	49	28	45.5
<i>Bromus diandrus</i>	7	6	7	24.8	33.2	32.3	9	11.8	7.8	12.8	10.7	12.3	15.7	10.7	15	13.8	10	10.3	51.3	43.2	45.5
<i>Bromus hordeaceus</i>	9	8	9	28.6	41.6	34.4	11.4	6.5	11	14	9.9	12.6	14.4	11.1	13.1	11	9.3	7	50.8	36.8	43.8
<i>Bromus lanceolatus</i>	4	2	4	29.5	42.5	40.5	12.5	6	9	13	12	13.5	14.5	10	11	9	7	11.5	49	35	45.5
<i>Bromus maritensis</i>	6	6	6	19	28.5	23	10	10.7	10.3	12.8	10.3	11.3	15.8	10.7	15.2	12.7	11.5	13.3	51.3	43.2	50.2
<i>Cynosurus echinatus</i>	1	1	1	54	67	58	11	7	10	14	5	8	12	5	10	5	4	7	42	21	30.5
<i>Dactylis glomerata</i>	4	4	4	55.8	56.5	52.8	9.5	11	9.8	13.5	7.3	10.3	13.3	9	12.8	7.5	7.8	5.8	43.8	35	38.5
<i>Elymus repens</i>	1	1	1	54	46	44	5	5	11	16	13	8	17	13	16	11	11	7	49	42	42
<i>Festuca arundinacea</i>	1	1	1	47	46	44	11	3	10	14	11	11	13	12	14	11	9	7	49	35	42
<i>Gaudinia fragilis</i>	2	1	2	54	60	65	12	4	4	13	8	8	12	9	9	5	7	7	42	28	28
<i>Hordeum geniculatum</i>	1	1	1	47	53	58	5	3	7	13	11	7	17	14	14	7	7	7	42	35	35
<i>Hordeum leporinum</i>	5	4	5	27.8	33.8	31.8	10.3	8.5	7.3	15.5	10.5	12	15.5	12.8	14.3	14.8	13.8	12	56	45.5	45.5
<i>Hyparrhenia hirta</i>	2	2	2	36.5	49.5	40.5	6	6	10.5	17	9.8	12.5	21	8	17.5	15.5	15	12	59.5	38.5	52.5
<i>Lolium rigidum</i>	5	2	4	36.5	53	47.5	8	8	6	13.5	8.5	9.5	17	9.5	16	10.5	12.5	10.5	49	38.5	42
<i>Stipa capensis</i>	5	5	5	47	53	45.4	8.8	5.8	8.2	10.2	7.8	10.6	12	8.4	11	11	8.8	6.6	42	30.8	36.4
<i>Taeniatherum caput-medusae</i>	3	3	3	47	53	46.3	7	5.3	7.3	10.3	9	11.7	14.3	7	10.7	10.3	9	7.7	42	30.3	37.3
<i>Trisetaria panicea</i>	6	3	5	51.7	46	55.7	9	8.7	7	11.7	8	10	12	7.3	10.3	7	8.7	7.7	39.7	32.7	35
<i>Vulpia geniculata</i>	7	7	7	42	52	38	11.9	6.7	12.3	13.3	8.1	12.4	14.7	9.9	14.7	10.1	8.3	10.6	50	33	50
<i>Vulpia myuros</i>	6	6	6	41.2	44.8	41.7	10	6.3	9.7	13.5	9.7	10.2	13.3	8	13.8	11	8.7	9.5	47.8	32.7	43.2

V: number of days elapsing from March 1st to the start of Phase 0, Phase 0: Preflowering Phase, Phase 1: Start of flowering, Phase 2: Full flowering, Phase 3: End of flowering, Range: Sum of the different phase lengths

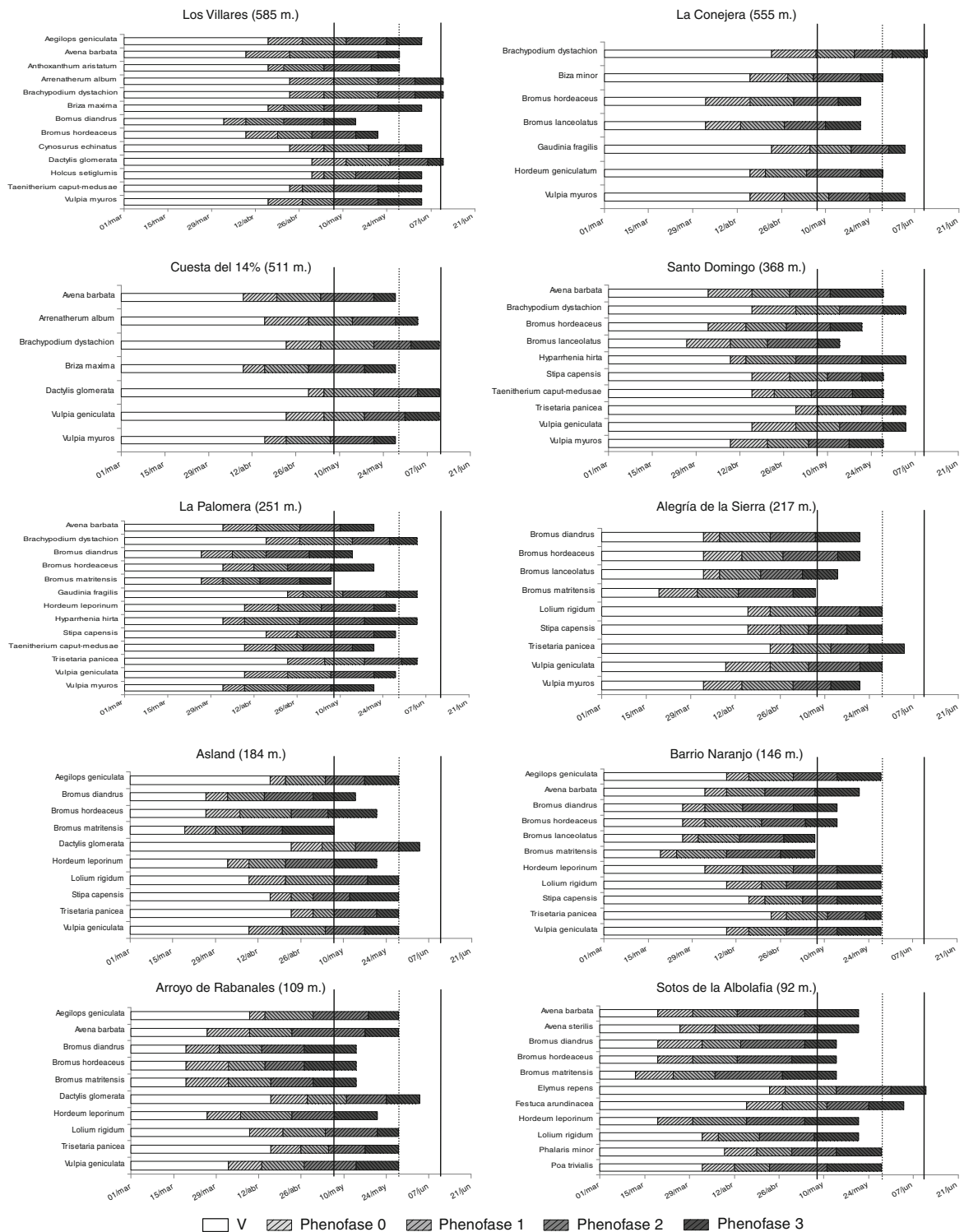


Fig. 6 Phenophase length in different populations in 2004. V: number of days elapsing from March 1st until the beginning of Phase 0, Phase 0: Preflowering Phase, Phase 1: Start of

flowering, Phase 2: Full flowering, Phase 3: End of flowering, Range: Sum of the different phase lengths

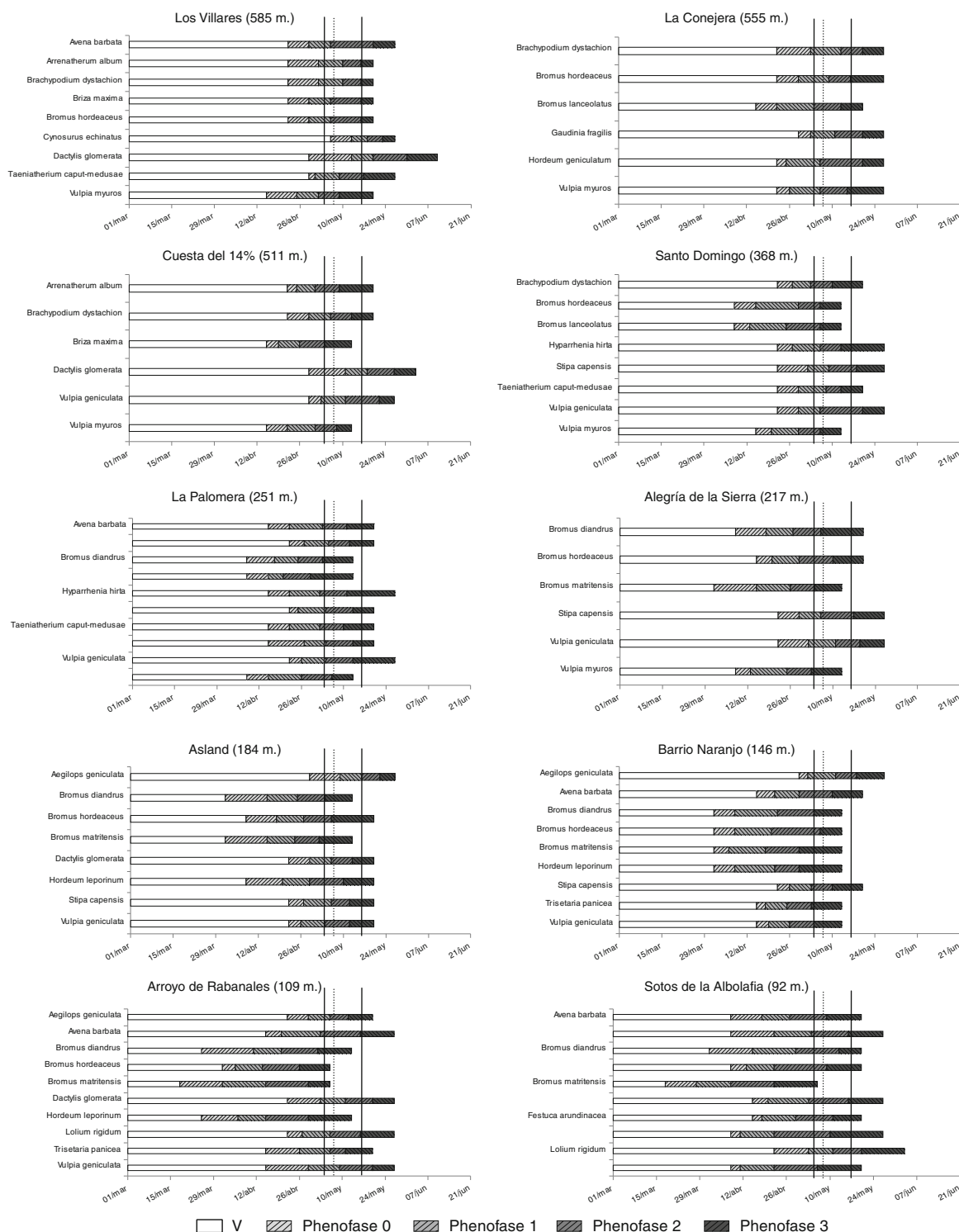


Fig. 7 Phenophase lengths in different populations in 2005. V: number of days elapsing from March 1st until the beginning of Phase 0, Phase 0: Preflowering Phase, Phase 1: Start of

flowering, Phase 2: Full flowering, Phase 3: End of flowering, Range: Sum of the different phase lengths

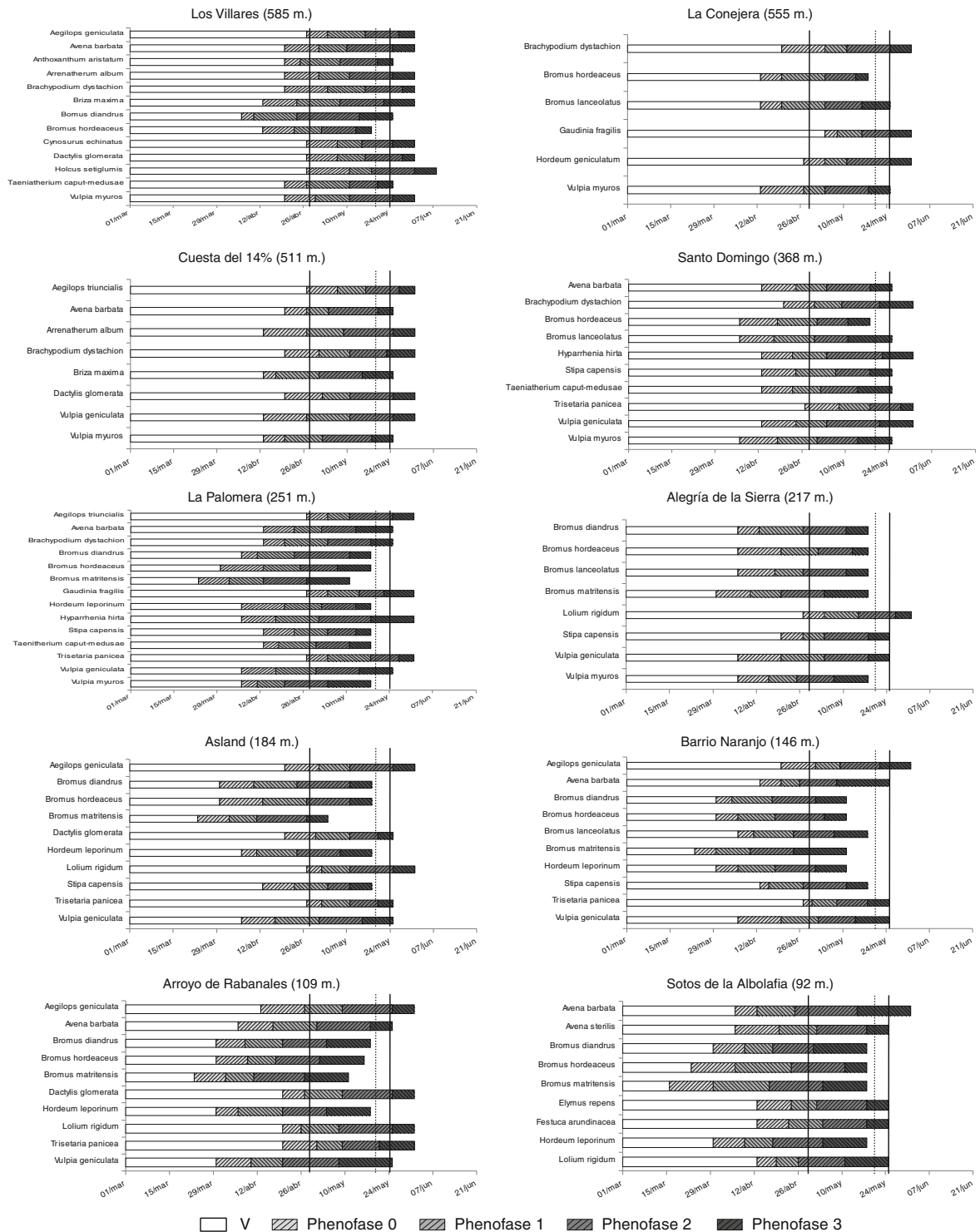


Fig. 8 Phenophase length in different populations in 2006. V: number of days elapsing from March 1st until the beginning of Phase 0, Phase 0: Preflowering Phase, Phase 1: Start of

flowering, Phase 2: Full flowering, Phase 3: End of flowering, Range: Sum of the different phase lengths

Table 4 Correlations between phenological and aerobiological data. 2004

	Spearman	<i>p</i> -level
<i>Brachypodium dystachion</i>		
La Palomera	0.692	0.003**
Santo Domingo	0.732	0.001**
Cuesta del 14%	0.769	0.000**
La Conejera	0.865	0.000**
Los Villares	0.860	0.000**
<i>Dactylis glomerata</i>		
Arroyo de Rabanales	0.693	0.003**
Asland	0.754	0.001**
Cuesta del 14%	0.862	0.000**
Los Villares	0.883	0.000**
<i>Lolium rigidum</i>		
Arroyo de Rabanales	0.694	0.003**
Asland	0.703	0.002**
Alegría de la Sierra	0.744	0.000**
Barrio Naranjo	0.669	0.005**
Sotos de la Albolafia	0.307	0.247
<i>Stipa capensis</i>		
Asland	0.447	0.082
Alegría de la Sierra	0.499	0.049*
Barrio Naranjo	0.461	0.073
La Palomera	0.529	0.035*
Santo Domingo	0.550	0.027*
<i>Trisetaria panicea</i>		
Arroyo de Rabanales	0.499	0.049*
Asland	0.587	0.017*
Barrio Naranjo	0.589	0.016*
La Palomera	0.780	0.000**
Alegría de la Sierra	0.726	0.001**
Santo Domingo	0.768	0.001**
<i>Vulpia geniculata</i>		
Arroyo de Rabanales	0.200	0.458
Asland	0.378	0.149
Alegría de la Sierra	0.445	0.084
Barrio Naranjo	0.326	0.218
La Palomera	0.433	0.094
Santo Domingo	0.721	0.002**
Cuesta del 14%	0.857	0.000**

* $p < 0.05$; ** $p < 0.01$

scarce rainfall registered during the survey season delayed the start of flowering, and species displayed their shortest phenological range, resulting in the

Table 5 Correlations between phenological and aerobiological data. 2005

	Spearman	<i>p</i> -level
<i>Brachypodium dystachion</i>		
La Palomera	0.804	0.000**
Santo Domingo	0.812	0.000**
Cuesta del 14%	0.800	0.000**
La Conejera	0.854	0.000**
Los Villares	0.794	0.000**
<i>Dactylis glomerata</i>		
Arroyo de Rabanales	0.857	0.000**
Asland	0.800	0.000**
Cuesta del 14%	0.810	0.000**
Los Villares	0.823	0.000**
<i>Lolium rigidum</i>		
Arroyo de Rabanales	0.872	0.000**
Sotos de la Albolafia	0.887	0.000**
<i>Stipa capensis</i>		
Asland	0.804	0.000**
Alegría de la Sierra	0.870	0.000**
Barrio Naranjo	0.801	0.000**
La Palomera	0.804	0.000**
Santo Domingo	0.867	0.000**
<i>Trisetaria panicea</i>		
La Palomera	0.772	0.001**
Santo Domingo	0.800	0.000**
Los Villares	0.827	0.000**
<i>Vulpia geniculata</i>		
Arroyo de Rabanales	0.860	0.000**
Asland	0.804	0.000**
Alegría de la Sierra	0.864	0.000**
Barrio Naranjo	0.622	0.013*
La Palomera	0.875	0.000**
Santo Domingo	0.868	0.000**
Cuesta del 14%	0.810	0.000**

* $p < 0.05$; ** $p < 0.01$

shortest pollen season and the lowest pollen counts. It should be noted that the phenological range of populations located at higher altitudes was shorter, because they flowered later and the scarce rainfall during May hindered their proper development.

As indicated by Orlandi et al. 2005, aerobiological and phenological methods can be used effectively as integrated research tools for this kind of study. Phenological data can be very useful in aerobiological

Table 6 Correlations between phenological and aerobiological data. 2006

	Spearman	p-level
<i>Brachypodium dystachion</i>		
La Palomera	0.738	0.002**
Santo Domingo	0.900	0.000**
Cuesta del 14%	0.905	0.000**
La Conejera	0.898	0.000**
Los Villares	0.899	0.000**
<i>Dactylis glomerata</i>		
Arroyo de Rabanales	0.902	0.000**
Asland	0.842	0.000**
Cuesta del 14%	0.905	0.000**
Los Villares	0.863	0.000**
<i>Lolium rigidum</i>		
Arroyo de Rabanales	0.901	0.000**
Asland	0.870	0.000**
Alegría de la Sierra	0.863	0.000**
Sotos de la Albolafia	0.684	0.005**
<i>Stipa capensis</i>		
Asland	0.596	0.019*
Alegría de la Sierra	0.841	0.000**
Barrio Naranjo	0.563	0.029*
La Palomera	0.596	0.019*
Santo Domingo	0.734	0.002**
<i>Trisetaria panicea</i>		
Arroyo de Rabanales	0.904	0.000**
Asland	0.826	0.000**
Barrio Naranjo	0.826	0.000**
La Palomera	0.866	0.000**
Santo Domingo	0.858	0.000**
<i>Vulpia geniculata</i>		
Arroyo de Rabanales	0.418	0.121
Asland	0.627	0.012*
Alegría de la Sierra	0.685	0.005**
Barrio Naranjo	0.595	0.019*
La Palomera	0.599	0.018*
Santo Domingo	0.805	0.000**
Cuesta del 14%	0.844	0.000**

* $p < 0.05$; ** $p\text{-level} < 0.01$

studies, because pollen grain release is related to flowering phenology and other environmental variables (Jato et al. 2002). Studies report positive correlations between phenological development and pollen counts in various tree genera, e.g. *Quercus* (Gómez Casero et al. 2007), *Ulmus* and *Fraxinus*

(Latorre and Bianchi 1998), *Betula* (Jato et al. 2002), *Alnus*, *Corylus* and *Betula* (Kasprzyk 2003) and *Cupressus* (Hidalgo et al. 2003). Phenological observations have also been used to confirm the real contribution of olive pollen to overall pollen curves (Orlandi et al. 2005; García-Mozo et al. 2006). In Córdoba, the pollen curve is characterised by a number of peaks attributable to the staggered pollination of various crops, sometimes located far from the pollen trap. In Perugia, the pollen curve is characterised by a single peak, because of the proximity of the crops studied (Fornaciari et al. 2000). Few studies have focussed on this correlation in herbaceous species, e.g. *Parietaria judaica* (Guardia and Belmonte 2004). It is essential to establish the phenological behaviour of different Poaceae species in order to determine which of them contribute most to the pollen curve in a given area. Only a few authors have drawn up lists of major local Poaceae species displaying a positive correlation between phenological development and pollen counts (Zerboni et al. 1991; Newham et al. 1995).

The results obtained here showed that, in all three study years, *Brachypodium dystachion*, *Dactylis glomerata*, *Lolium rigidum*, *Stipa capensis*, *Trisetaria panicea* and *Vulpia geniculata* displayed a strong correlation between full flowering period and main pollen season. Results also showed that populations located at higher altitudes contributed more to the Poaceae pollen curve. Strikingly, only four of the species in the study area (*Dactylis glomerata*, *Lolium rigidum*, *Trisetaria panicea* and *Vulpia geniculata*) are among the main pollen-producing Poaceae species (Prieto-Baena et al. 2003).

5 Conclusions

The floral development of the species studied is strongly influenced by various factors, including altitude, rainfall and temperature. Differences in the length of each phenophase and in total floral development were observed between different species, and also between different locations for the same species.

Taking into account both plant pollen production and floral phenology, *Dactylis glomerata*, *Lolium rigidum*, *Trisetaria panicea* and *Vulpia geniculata* would appear to be the species contributing most to the grass pollen curve, especially those populations located on the mountain slopes of Córdoba.

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