

Phenological records as a complement to aerobiological data

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Abstract Phenological studies in combination with aerobiological studies enable one to observe the relationship between the release of pollen and its presence in the atmosphere. To obtain a suitable comparison between the daily variation of airborne pollen concentrations and flowering, it is necessary for the level of accuracy of both sets of data to be as similar as possible. To analyse the correlation between locally observed flowering data and pollen counts in pollen traps in order to set pollen information forecasts, pollen was sampled using a Burkard volumetric pollen trap working continuously from May 1993. For the phenological study we selected the main pollen sources of the six pollen types most abundant in our area: Cupressaceae, *Platanus*, *Quercus*, *Plantago*, *Olea*, and Poaceae with a total of 35 species. We selected seven sites to register flowering or pollination, two with semi-natural vegetation, the rest being urban sites. The sites were visited weekly from March to June in 2007, and from January to June in 2008 and 2009. Pollen shedding was checked at each visit, and recorded as the percentage of flowers or microsporangia in that state. There was an association between flowering phenology and airborne pollen records for some of the pollen types (*Platanus*, *Quercus*, *Olea* and *Plantago*). Nevertheless, for the other types (Cupressaceae and Poaceae) the flowering

and airborne pollen peaks did not coincide, with up to 1 week difference in phase. Some arguments are put forward in explanation of this phenomenon. Phenological studies have shown that airborne pollen results from both local and distant sources, although the pollen peaks usually appear when local sources are shedding the greatest amounts of pollen. Resuspension phenomena are probably more important than long-distance transport in explaining the presence of airborne pollen outside the flowering period. This information could be used to improve pollen forecasts.

Keywords Aerobiology · Allergy · Forecasting · Phenology · Pollen · Pollen trap

Introduction

Phenology is the study of periodic plant and animal life cycle events and how these are influenced by seasonal and yearly variations in weather. Phenophase is an observable stage or phase in the annual life cycle of a plant or animal that can be defined by a start and an end point. Phenophases generally have lengths of a small number of days or weeks. Examples include the period over which newly emerging leaves are visible, or the period over which open flowers are present on a plant.

The presence of pollen in the atmosphere follows a clear seasonal model in response to the seasonal flowering of the plant sources. Pollen captured from the air is difficult to identify at species level unless there is only a single species with a distinctive morphological pollen grain as the source. In aerobiology, therefore, the term pollen type is used. This can include various species, genera, or even families. Phenological studies allow one to follow pollen release

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from the sources at species level by determining the flowering interval and peak for each species, and to attribute the responsibility for allergic symptoms to a particular species (Zerboni and Manfredi 1988; Ickovic et al. 1989; Orlandi et al. 2005). The core interest in these areas of phenological research lies in their importance for the study of climate change (Schwartz 1998). In the present case, however, we use this information to elaborate and send by SMS (short message service) short reports about the main airborne pollen types in the atmosphere and their forecast, as a free public service offered by the University of Extremadura to people interested in airborne pollen concentration.

While one would naturally expect there to be a correspondence between the shedding of pollen and its presence in the air, in only a few cases, such as in Cupressaceae (Hidalgo et al. 2003), has that correspondence been observed. Wind modifies this relationship by transporting the pollen far from its source to the site being studied aerobiologically. Hence, not all the pollen grains captured can be explained by flowering of local vegetation, as has been shown for *Betula* pollen in Finland (Ranta et al. 2006), Denmark (Skjøth et al. 2007), and Spain (Jato et al. 2007a), for *Quercus* pollen (Jato et al. 2007b), and for other pollen types (Estrella et al. 2006) in Germany. It is thus important for phenological studies to include all the possible sources of pollen in the air, or for forecasting to take into account the main atmospheric transport models. Indeed, since much pollen may be captured before and after the flowering period, in some cases modelling has to integrate aerobiological and phenological data over an extensive geographical range, for example regional data of an entire country (García-Mozo et al. 2008).

Phenological studies related to aerobiology can be performed at a widely varying range of detail. Data can be gathered from herbaria or literature sources (Fairley and Batchelder 1986; Lorenzoni et al. 1998), although in these cases the frequency of observations is usually low, and only the start and end dates of flowering are used (Ickovic et al. 1989). Some studies have used the three levels of flowering intensity defined by Marcello (1935) or very similar criteria (Latorre 1997; Kasprzyk 2003). For other purposes, more phenophases can be defined, mainly reflecting the plants' reproductive biology: 5 levels in Cupressaceae (Hidalgo et al. 2003), 7 levels in Poaceae (Zanotti and Puppi 2000), and 12 reproductive phenophases in the olive (Fornaciari et al. 2000). The most widely accepted frequency for phenological observations is weekly, although in some cases this is increased to twice weekly (Hidalgo et al. 2003; Fornaciari et al. 2000; Latorre 1999). While counting the flowers on each plant is tedious and time consuming, it provides highly valuable data. This method is usually applied to some randomly selected specimens of each

species, or for different levels with randomly selected plants, branches, inflorescences, and flowers (Latorre 1997, 1999).

In phenological studies it is very important to verify that the flowers are shedding pollen (since this is the phenophase responsible for the presence of airborne pollen), and not just to count opened flowers because these might have already released all their pollen. Nevertheless, pollen is released by only some flowers at the beginning and at the end of the phenophase and with full emission between both. Pollen shedding usually occurs only at certain hours of the day, and depends directly on the weather conditions. Usually, this state of full emission is evaluated when all the stamens are extruded (Zanotti and Puppi 2000). Qualitative phenological information can be transformed into quantitative data to obtain daily averages of flowering plant percentages for each pollen type, and these data can be compared with pollen concentrations (Orlandi et al. 2005).

The aim of the present study was to analyse the correlation between locally observed flowering data and pollen trap counts in order to provide pollen information forecasts.

Material and methods

Aerobiological study

Pollen was sampled using a 7-day Burkard volumetric pollen trap (Hirst 1952) that has been operating continuously since May 1993. It is located in the Agrarian Engineering School of the University of Extremadura in Badajoz (SW Spain, latitude 38.89 longitude -6.97), on a roof terrace at 6 m above ground level. The samples obtained were read using the method recommended by the Spanish Aerobiology Network (REA; Domínguez et al. 1992). The result is a daily average expressed as pollen grains/m³. To compare these data with phenological data, they have been weekly averaged from 17 weeks in 2007 (from March to June) and 26 weeks in 2008 and 2009 (from January to June). The climate of the study site is Mediterranean, with an annual average temperature of 16.4°C and 483.4 mm of annual rain (1961–1990 period, data from the Regional Meteorological Centre). Winds are nearly constantly from the west or south–west.

Phenological study

For the phenological study, the main pollen sources of the six most abundant pollen types in our area were selected, comprising a total of 35 species: Cupressaceae pollen type (*Cupressus sempervirens*, *C. arizonica*, *C. macrocarpa*, *Platyclusus orientalis*, *Calocedrus decurrens*), *Platanus*

pollen type (*Platanus hispanica*, *P. orientalis*), *Quercus* pollen type (*Quercus rotundifolia*, *Q. suber*), *Plantago* pollen type (*Plantago coronopus*, *P. lagopus*), *Olea* pollen type (*Olea europaea*), and Poaceae pollen type (*Agrostis castellana*, *A. pourretii*, *Avena barbata*, *A. sterilis*, *Brachypodium distachyon*, *Bromus diandrus*, *B. hordeaceus*, *Cynodon dactylon*, *Cynosurus echinatus*, *Dactylis glomerata*, *Gaudinia fragilis*, *Hordeum leporinum*, *Lamarckia aurea*, *Lolium rigidum*, *Molineriella minuta*, *Phalaris minor*, *Piptatherum miliaceum*, *Poa annua*, *Rostraria cristata*, *Sorghum halepense*, *Stipa capensis*, *Trisetum paniceum*, *Vulpia geniculata*, *V. myuros*).

To observe flowering or pollination, we selected seven sites (Table 1, Fig. 1), two (A, B) with semi-natural vegetation (holm oak, *Quercus rotundifolia*, and cork oak, *Quercus suber*, trees with Mediterranean shrubs), and the rest (C–G) urban sites with ornamental shrubs and trees and nitrophilous herbs. The sites were visited weekly on average from March to June in 2007, and from January to June in 2008 and 2009. Pollen shedding was verified on each visit by touching or shaking the inflorescences or male cones on dry sunny days at solar midday, and then recording the percentage of flowers or microsporangia in this state. For trees (*Quercus*, *Olea*, *Platanus*, and Cupressaceae), three specimens were selected at each site, and ten regularly distributed branches around the entire cup were selected, to include any possible variation due to orientation as exposure to sunlight. For the herbaceous species (Poaceae and *Plantago*), some areas of about 1 m² including one or more species were selected to complete all

the species present and ten inflorescences were selected in each area for each species. For some of the grasses that usually had no open spikelets (*Bromus diandrus*, *B. hordeaceus*, *Hordeum leporinum*, *Vulpia myuros*), we took into account the inflorescences present just after the opening of the flag leaf.

Statistical analyses

To analyse the association between airborne pollen and phenological data, the Spearman correlation coefficient was calculated between the weekly pollen concentrations for the day of the phenological study and that day's phenological data. For some of the pollen types with various species (Cupressaceae, *Quercus*, Poaceae and *Plantago*), the sum of the phenological data for all the species involved was used.

Results

Figures 2, 3, 4, 5, 6, 7, 8, and 9 show the phenological results compared with the airborne pollen levels for the three years studied, two for Cupressaceae. In chronological order of flowering, they are as follows:

The Cupressaceae family is represented by five species in the study area. The most frequent are *Cupressus sempervirens* (44%), *Platyclusus orientalis* (28%), and *Cupressus arizonica* (20%). The other two species, *Cupressus macrocarpa* and *Calocedrus decurrens*, repre-

Table 1 Places and species selected for the phenological study in Badajoz (Spain), see Fig. 1

Place	Species
A. Tres Arroyos area (Corte de Peleas road BA-022) latitude 38.844788 longitude -6.884651	<i>Agrostis castellana</i> , <i>A. pourretii</i> , <i>Avena barbata</i> , <i>Bromus diandrus</i> , <i>B. hordeaceus</i> , <i>Dactylis glomerata</i> , <i>Gaudinia fragilis</i> , <i>Hordeum leporinum</i> , <i>Lamarckia aurea</i> , <i>Lolium rigidum</i> , <i>Molinerilla minuta</i> , <i>Plantago coronopus</i> , <i>Phalaris minor</i> , <i>Quercus rotundifolia</i> , <i>Stipa capensis</i> , <i>Trisetum paniceum</i> , <i>Vulpia geniculata</i> , <i>V. myuros</i>
B. Finca Las Arenosas (Olivenza road EX-107) latitude 38.791289 longitude -7.049618	<i>Avena barbata</i> , <i>A. sterilis</i> , <i>Bromus diandrus</i> , <i>B. hordeaceus</i> , <i>Cynodon dactylon</i> , <i>Cynosurus echinatus</i> , <i>Hordeum leporinum</i> , <i>Lamarckia aurea</i> , <i>Lolium rigidum</i> , <i>Molinerilla minuta</i> , <i>Phalaris minor</i> , <i>Plantago coronopus</i> , <i>P. lagopus</i> , <i>Piptatherum miliaceum</i> , <i>Quercus rotundifolia</i> , <i>Q. suber</i> , <i>Rostraria cristata</i> , <i>Trisetum paniceum</i> , <i>Vulpia geniculata</i> , <i>V. myuros</i>
C. La Granadilla (urban area) latitude 38.854614 longitude -7.001638	<i>Avena barbata</i> , <i>A. sterilis</i> , <i>Bromus diandrus</i> , <i>B. hordeaceus</i> , <i>Cynodon dactylon</i> , <i>Hordeum leporinum</i> , <i>Lamarckia aurea</i> , <i>Lolium rigidum</i> , <i>Olea europaea</i> , <i>Phalaris minor</i> , <i>Piptatherum miliaceum</i> , <i>Platanus hispanica</i> , <i>Poa annua</i> , <i>Sorghum halepense</i> , <i>Stipa capensis</i> , <i>Trisetum paniceum</i> , <i>Vulpia geniculata</i> , <i>V. myuros</i>
D. River walk, La Molineta garden (urban area) latitude 38.8762 longitude -6.986275	<i>Cupressus sempervirens</i> , <i>C. arizonica</i> , <i>Platyclusus orientalis</i> , <i>Calocedrus decurrens</i>
E. La Alcazaba castle (urban area) latitude 38.882748 longitude -6.970224	<i>Olea europaea</i>
F. Montero de Espinosa street (urban area) latitude 38.864172 longitude -6.97679	<i>Platanus orientalis</i>
G. University campus (urban area) latitude 38.887626 longitude -7.011337	<i>Cupressus macrocarpa</i>

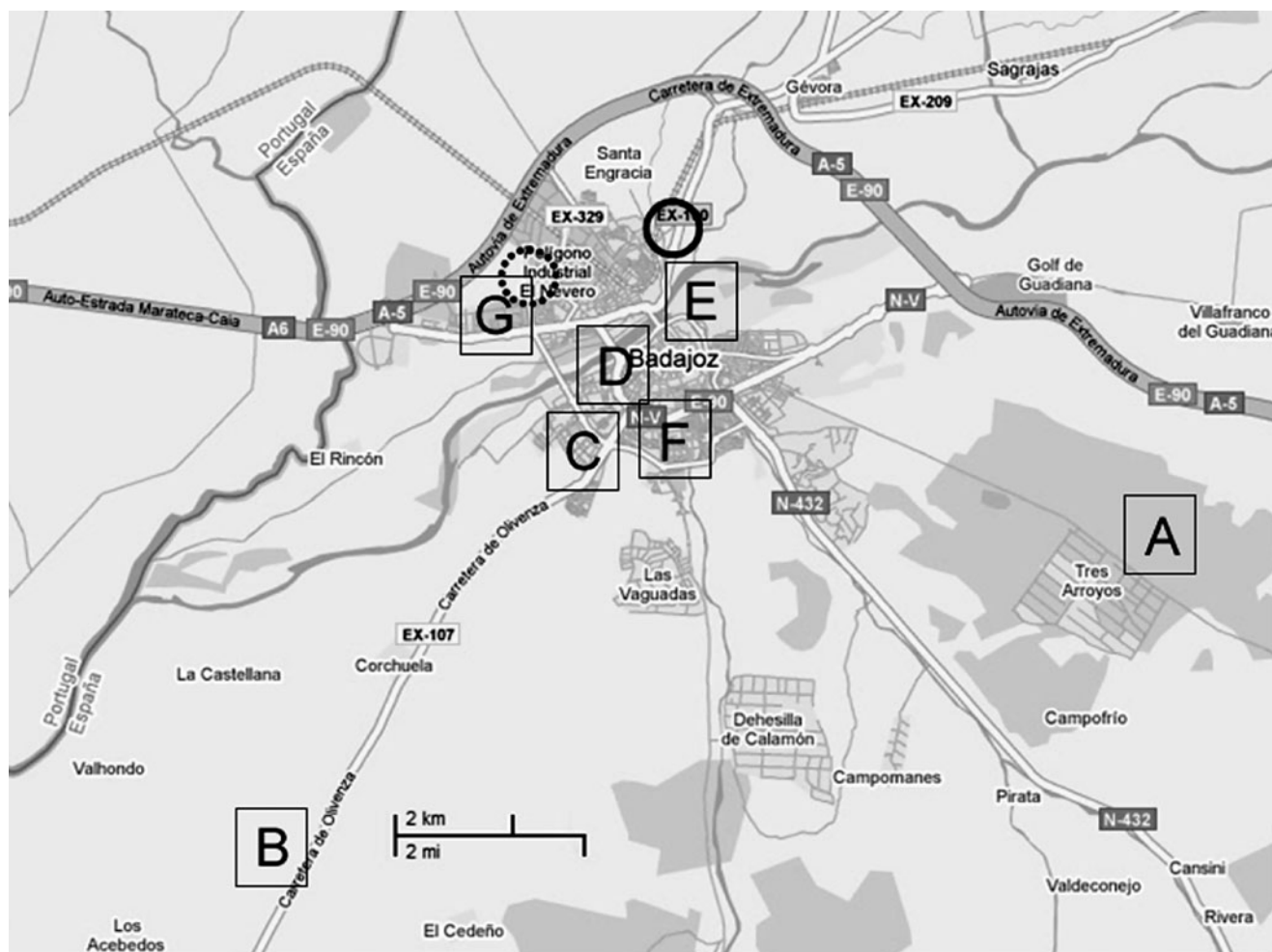


Fig. 1 Map of the city of Badajoz and the sites studied phenologically (see Table 1). Full circle Sampler at the Agrarian Engineering School, dotted circle sampler ant Science Faculty

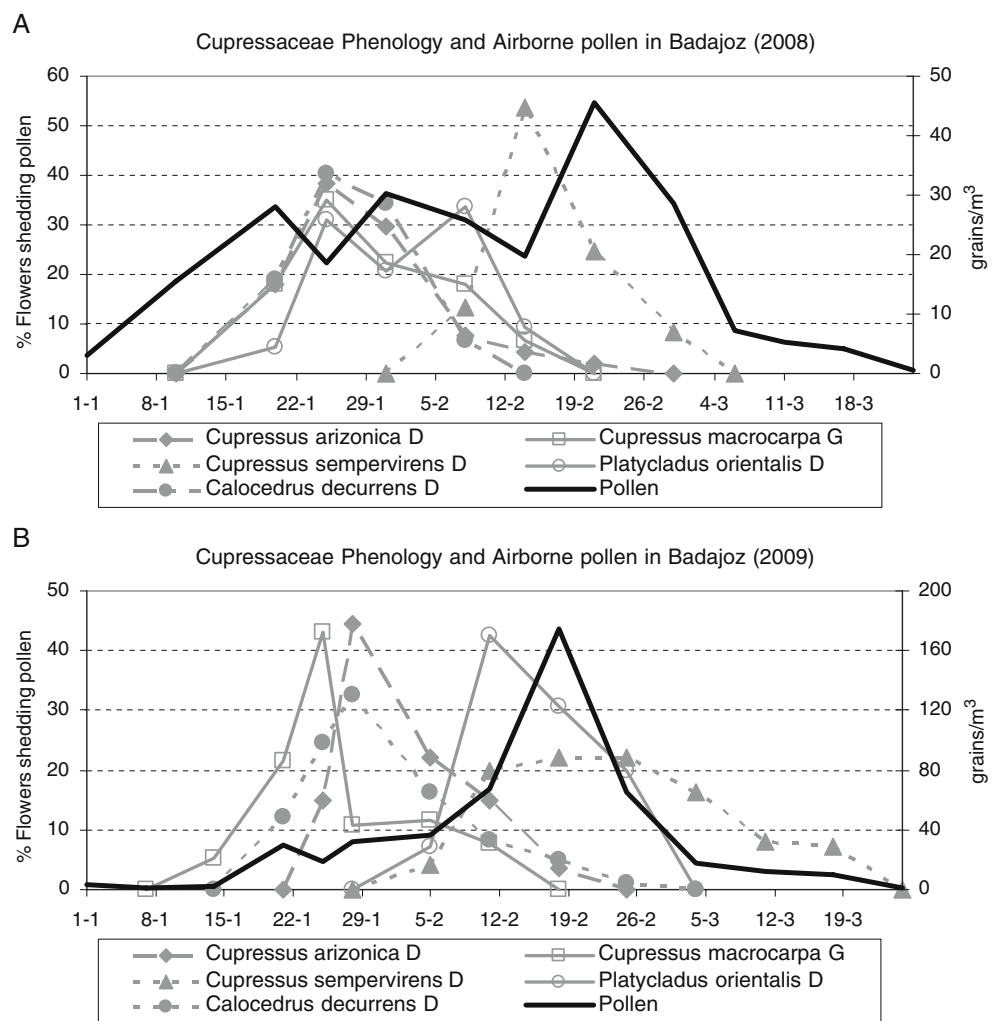
sent less than 8%. These species are widely spread in the city. In the 1st study year, pollen appeared in the atmosphere about 10 days before the onset of pollination, and remained for at least 2 weeks after pollination, but in the 2nd year there was hardly any pollen outside the pollination period (Fig. 2).

Two species of *Platanus* are present as ornamentals. The most abundant is *P. hispanica*, about twice as frequent in number as *P. orientalis*. Phenological peaks in flowering were recorded at the same time as airborne pollen peaks (Fig. 3). In the 1st study year, pollen was detected in the atmosphere about 1 week in advance of observed flowering, but in the 2nd and 3rd years there was no such delay. After flowering, only a few airborne pollen grains were recorded. In the 1st year, the two species showed very similar flowering patterns, but in the 2nd and 3rd year, *P. orientalis* was advanced by about 10–15 days. In the 2nd and 3rd year, the pollen peak occurred about 15 days earlier than in the 1st year.

The two species of *Quercus* studied (Fig. 4) have flowering phenophases separated by about 20–30 days, with *Q. rotundifolia* flowering first and then *Q. suber*, so that when *Q. rotundifolia* flowering finishes *Q. suber* flowering has just started. Their representation in the vegetation is very different: *Q. rotundifolia* is widespread, while *Q. suber* is restricted to the wettest sites or mountainous zones, and never grows on calcareous soils. Thus, most of the airborne pollen recorded belonged to *Q. rotundifolia*. There was an apparent close coincidence between the airborne pollen peaks and the flowering peaks. Flowering started a few (5–10) days before the airborne pollen was recorded, and was present in the atmosphere for up to more than 1 month after flowering ended. In the 2nd year, the pollen peak appeared nearly 20 days earlier than in the 1st year and nearly 30 days in the 3rd year.

The olive has a short pollination period and a well-defined pattern of flowering. Pollen appears in the air about (0)5–7 days before flowering starts (Fig. 5). The peaks were

Fig. 2 Pollination phenology and airborne pollen concentration of Cupressaceae in 2008 (a) and 2009 (b)



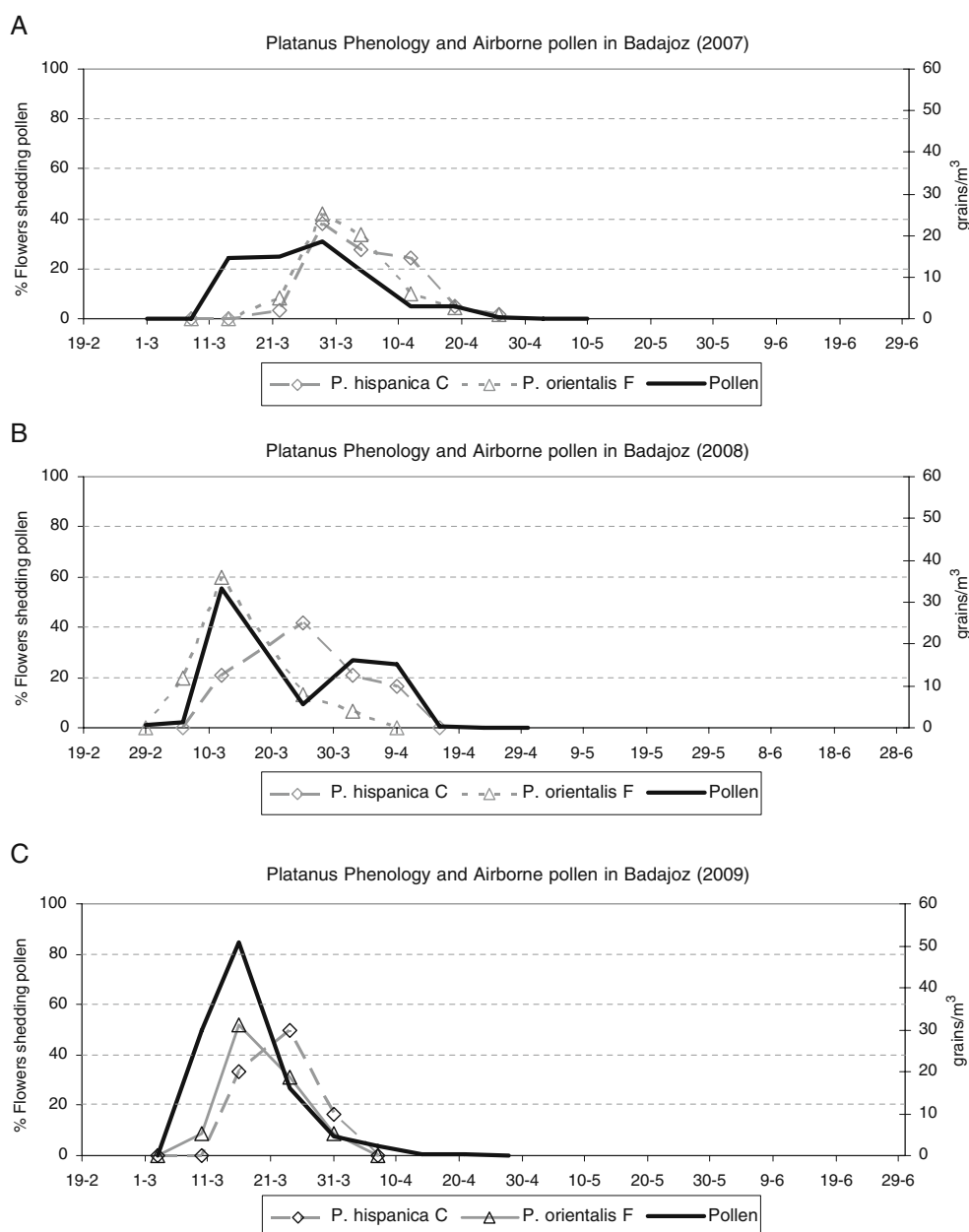
separated by 1 week in the 1st year, but were coincident in the 2nd and the 3rd year. There was a long period (more than 1 month) with pollen in the atmosphere after flowering had finished, but the onset of flowering was nearly at the same time as the beginning of airborne pollen records. In the 2nd and 3rd year, there was an advance in the pollen peak relative to the first of about 10–15 days.

Grass species were studied phenologically at sites A, B, and C (Fig. 1), two of which correspond to semi-natural vegetation: holm oak (site A) and cork oak (site B), and one to an urban area (site C). The total number of species was 24, but the number was different at each site even in each year (Table 2). Five species are perennial, and the rest annual. Only three species appeared at all three sites studied (*Hordeum leporinum*, *Trisetum paniceum*, and *Vulpia geniculata*). Some species were characteristic of one particular site (*Agrostis castellana* and *Agrostis pourretii* at site A, and *Cynosurus echinatus* at site B). Some annual species appeared in sufficient quantity for phenological counting only in one year, mainly due to the meteorological conditions (abundance of rainfall). This was the case for

Brachypodium distachyon and *Rostraria cristata*. The only perennial plant presents in one of the years was *Sorghum halepense*, because its presence was so low at the site selected that in the 2nd and 3rd year its contribution was insignificant. Figures 6–8 show the accumulated contribution of species implied in each site, so more than 100% is reached. In 2007 pollen concentration was higher than in 2008 and 2009 as a consequence of higher autumn–winter rain in the first year studied.

For site A, it was possible to separate two groups of species based in their flowering phenology. The first group included *Avena barbata*, *Brachypodium distachyon*, *Bromus diandrus*, *Bromus hordeaceus*, *Hordeum leporinum*, *Lamarckia aurea*, *Molineriella minuta* and *Stipa capensis*, flowering mainly in March. The second group included *Agrostis pourretii*, *Agrostis castellana*, *Dactylis glomerata*, *Gaudinia fragilis*, *Lolium rigidum*, *Phalaris minor*, *Trisetum paniceum*, *Vulpia geniculata*, and *Vulpia myuros*, flowering mainly at the end of April and beginning of May. The first group of species contributed hardly at all to the pollen in the atmosphere; it was the second group that was responsible for

Fig. 3 Flowering phenology and airborne pollen concentration of *Platanus* species in 2007 (a), 2008 (b) and 2009 (c)



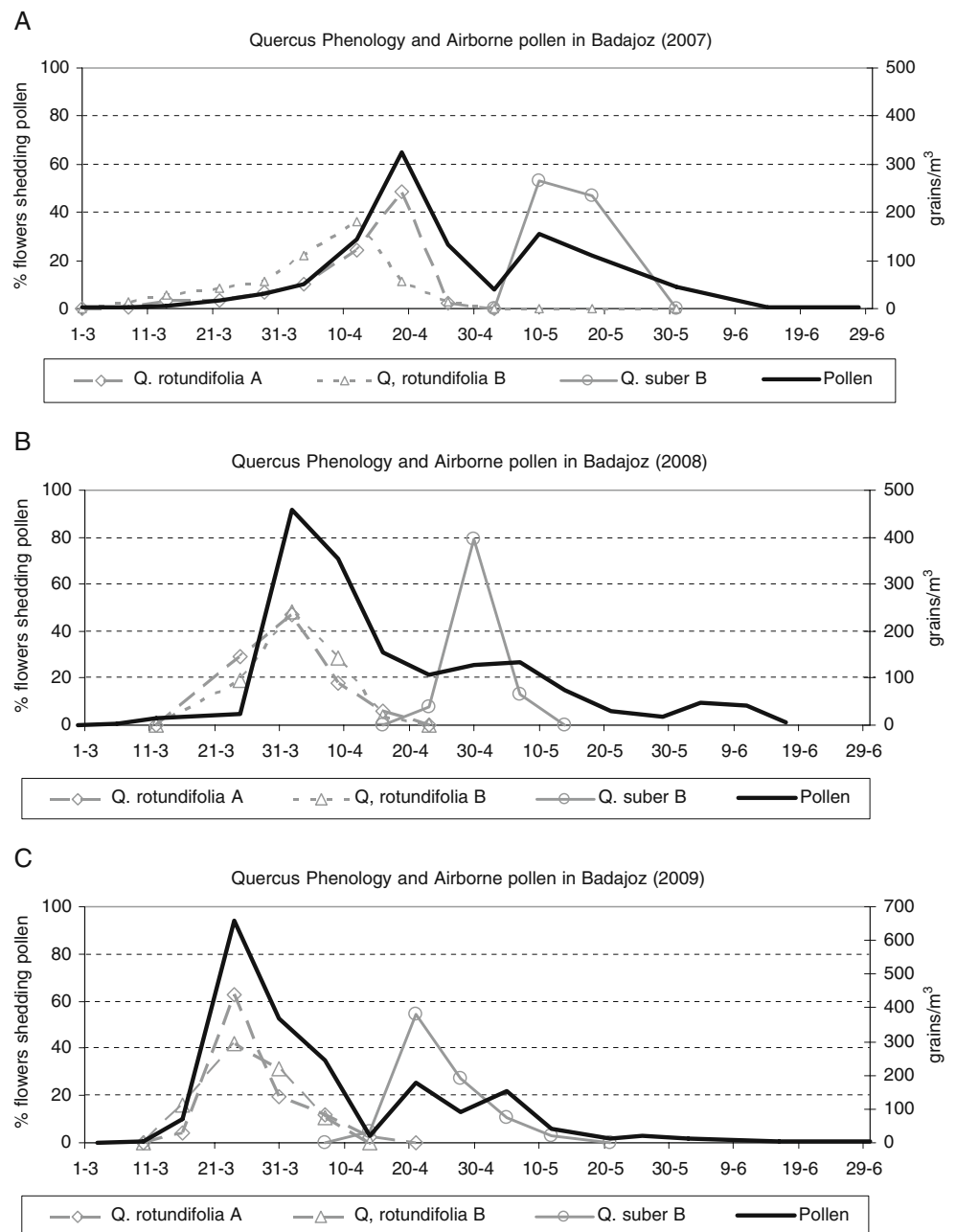
the airborne pollen. Nevertheless, there was a clear absence of correspondence between the flowering peak and the pollen peak. For site A, the first phenological peak appeared about 10 days before the pollen peak in the 1st year, in the 2nd year they occurred on the same dates, and in the 3rd year it was again very similar to the first one. For site B, the 2nd year's phenological peak occurred about 15 days before the 1st year's, with an additional two lower peaks, and in the 3rd year the phenological peak occurred 10 days earlier than in the 1st. For site C, in the 2nd year the phenological peak was advanced by about 25 days with respect to the 1st year, and the 3rd year the advance was by 10 days.

The *Plantago* species studied, *P. coronopus* and *P. lagopus*, showed great variability in their airborne presence

from year to year, probably reflecting different weather conditions. Their flowering phenophases overlapped, with no clear differentiation possible (Fig. 9). Airborne pollen was recorded about 10–15 days after the onset of flowering, and continued for 1–2 weeks after it had ended. It seems that *P. lagopus* was principally responsible for the pollen in the atmosphere because the pollen peaks coincided with its flowering peaks. Furthermore, it was also confirmed to be more abundant in the area than *P. coronopus*. The observed phenological features seemed to occur earlier in the 2nd and 3rd year with respect to the 1st.

The results of the correlation analysis between airborne pollen and phenological data are presented in Table 3. Except for Cupressaceae and Poaceae in 2008, the

Fig. 4 Flowering phenology and airborne pollen concentration of *Quercus* species in 2007 (a), 2008 (b) and 2009 (c)



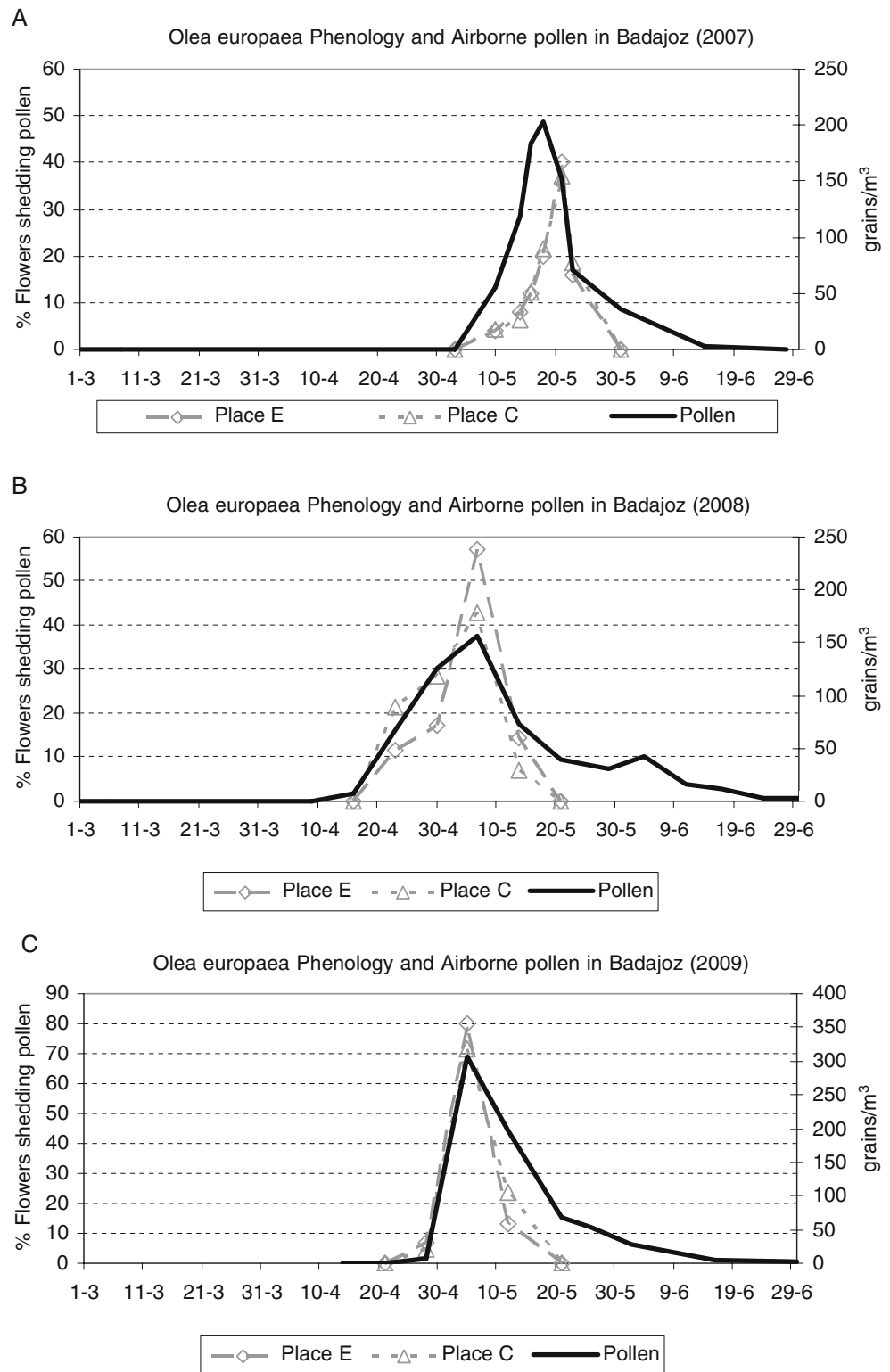
correlations were statistically significant in all studied years.

Discussion

Our results confirmed observations that the flowering phenology of sources near an aerobiological sampling site does not precisely match the airborne pollen records, although there was a statistically significant correlation between the variations in the two sets of data in nearly all cases. While an increase of sample size would provide more accurate results, including microclimatic variations,

correlation analysis showed that number of trees observed and places counted seem to be sufficient for the aims of this work. The environment around the city is quite homogeneous, there are no mountains or geographical anomalies, except for the vegetation of the river Guadiana, which does not include the main sources of airborne pollen. That airborne pollen appears both before and after the shedding of pollen was not evident in all the present cases. For the trees studied, there was always pollen in the atmosphere after flowering but not always before, as was the case in *Platanus* and *Olea* in 2007 and Cupressaceae in 2008. These cases could reflect the near absence of other pollen sources apart from those in the urban area since they are all

Fig. 5 Flowering phenology and airborne pollen concentration of *Olea europaea* in 2007 (a), 2008 (b) and 2009 (c)



non-natural, planted trees. Also, for the herbaceous pollen types studied—grasses and *Plantago*—there was no airborne pollen before flowering.

For the Cupressaceae family, there was no correspondence between phenology and airborne pollen data, in agreement

with the findings of Hidalgo et al. (2003), who found *Cupressus macrocarpa* to be the main pollen source in their study. In our case, it was *C. sempervirens*, which is most extensively planted as an ornamental (Gonzalo et al. 2006), and there was a correspondence between the airborne pollen

Fig. 6 Flowering phenology and airborne pollen concentration of Poaceae species at site A for 2007 (a), 2008 (b) and 2008 (c). Sums for all the species at site A (see Table 2)

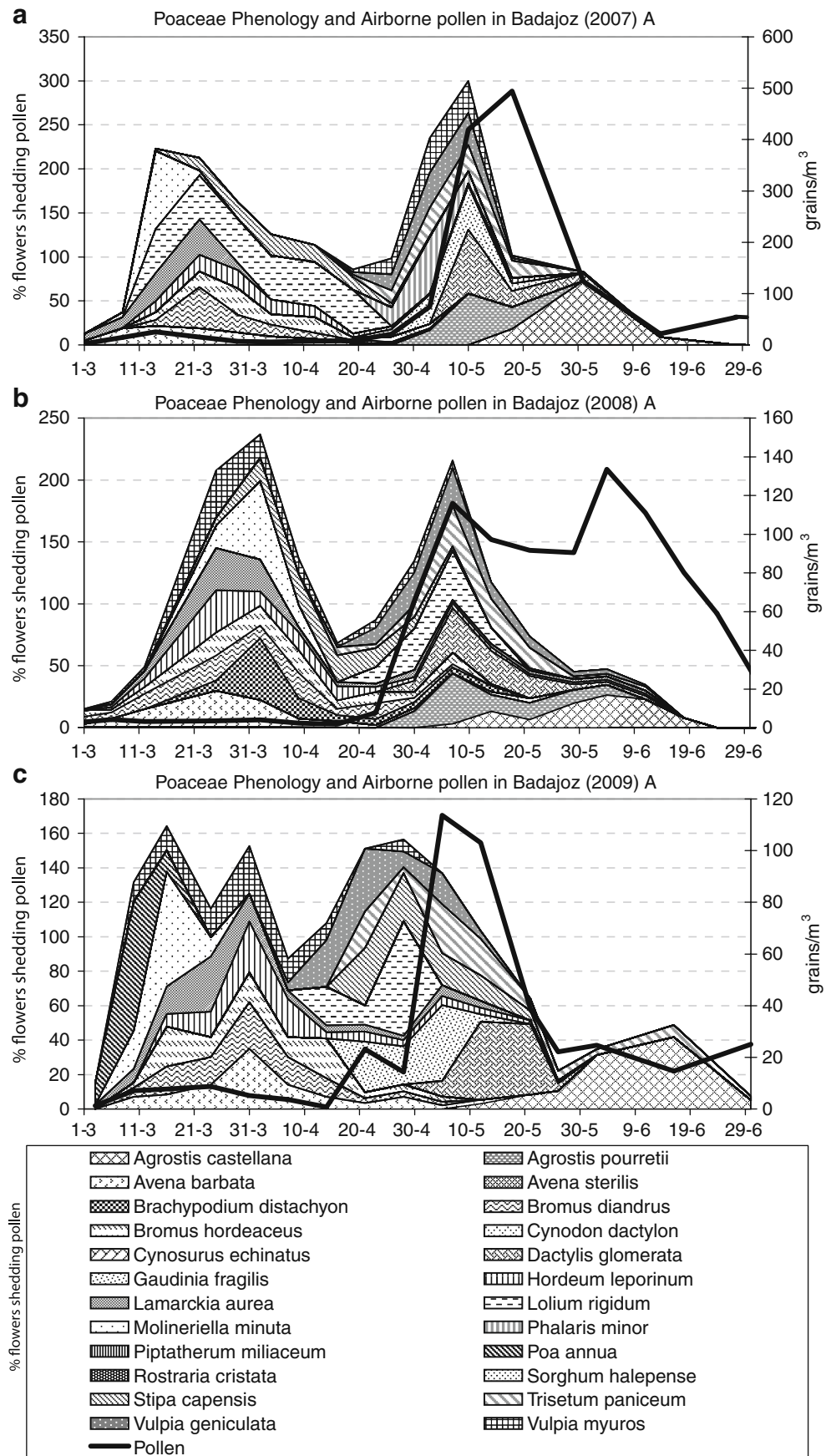


Fig. 7 Flowering phenology and airborne pollen concentration of Poaceae species at site B for 2007 (a), 2008 (b) and 2008 (c). Sums for all the species at site B (see Table 2)

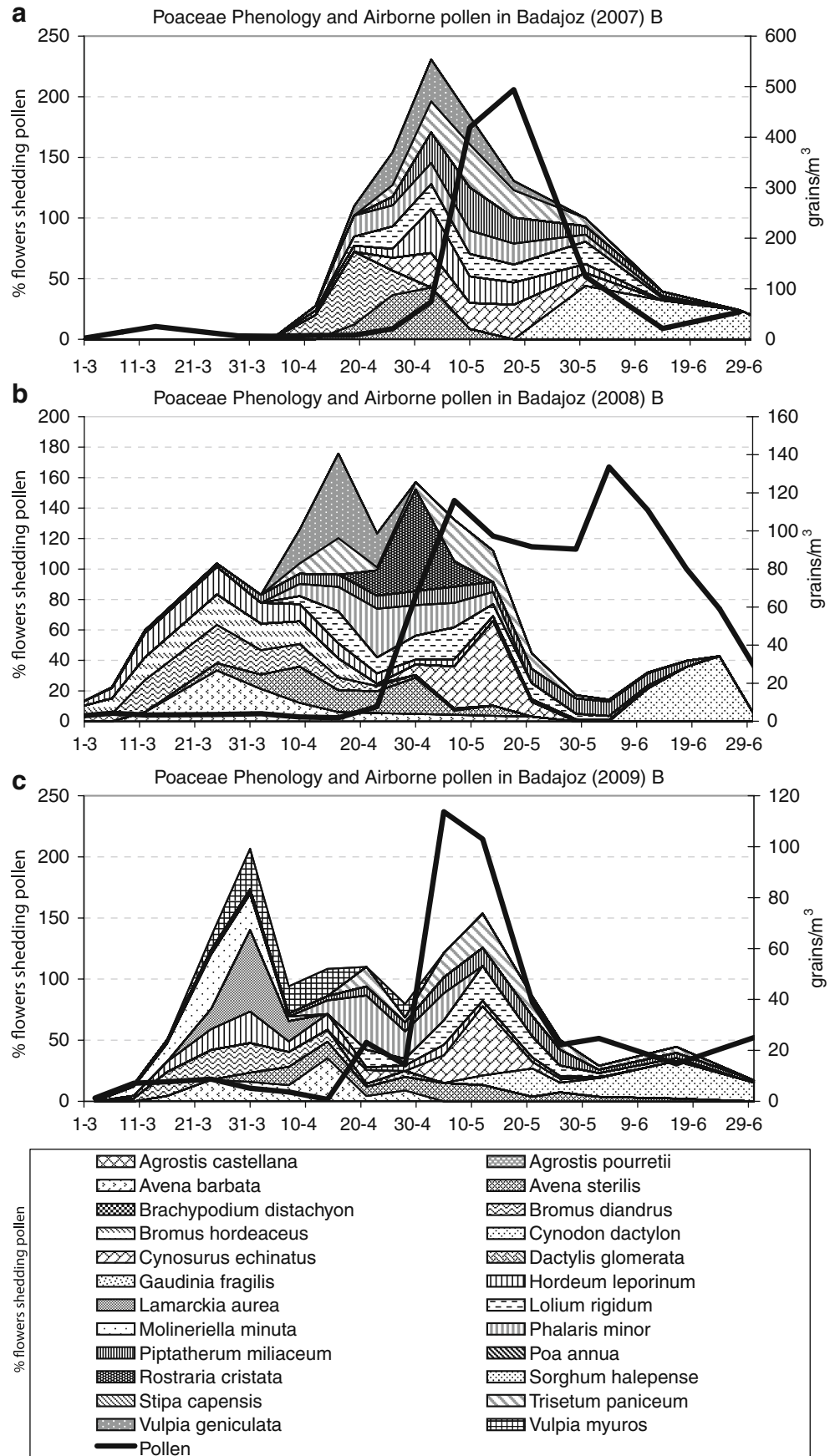


Fig. 8 Flowering phenology and airborne pollen concentration of Poaceae species at site C for 2007 (a), 2008 (b) and 2008 (c). Sums for all the species at site C (see Table 2)

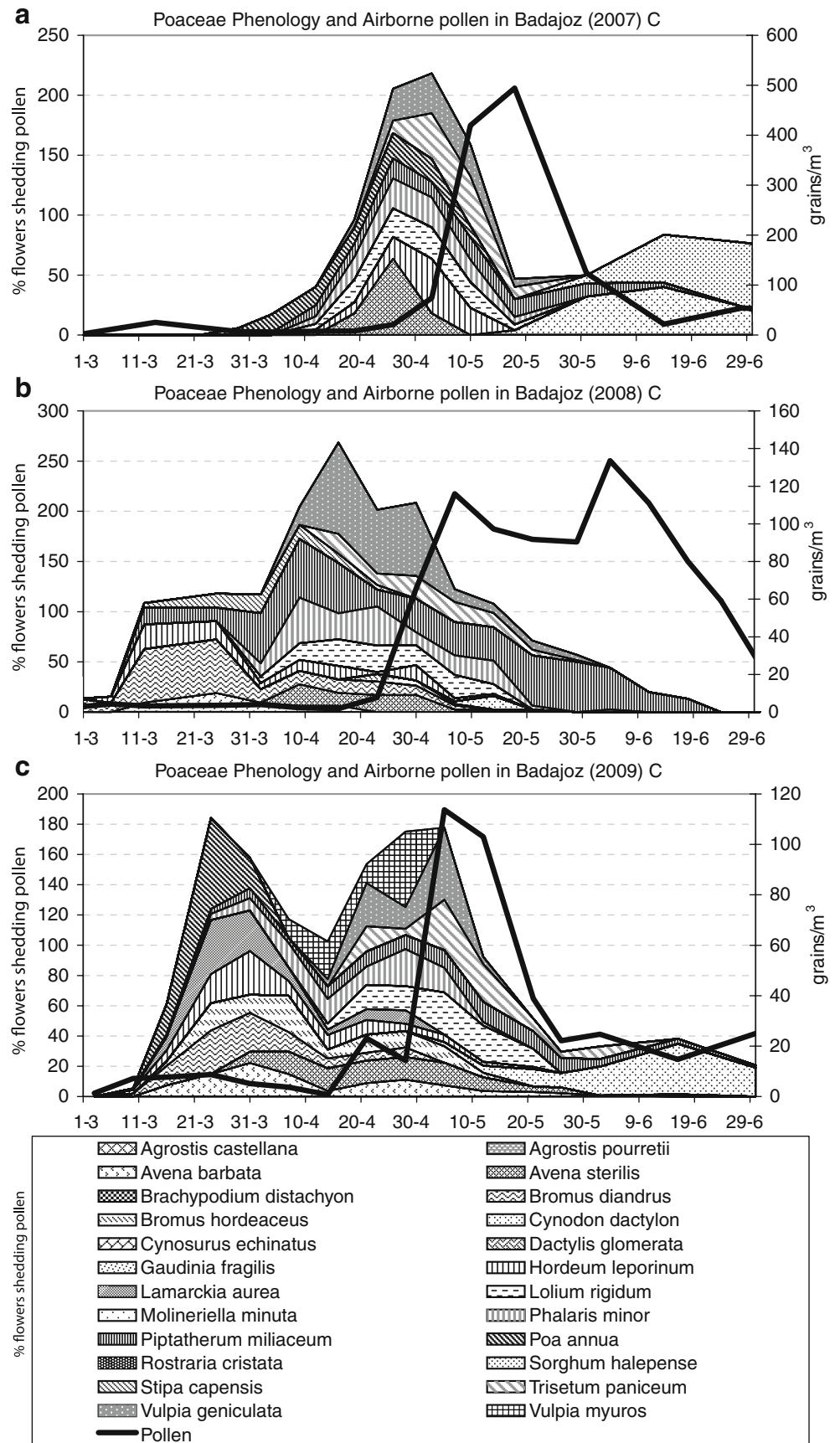
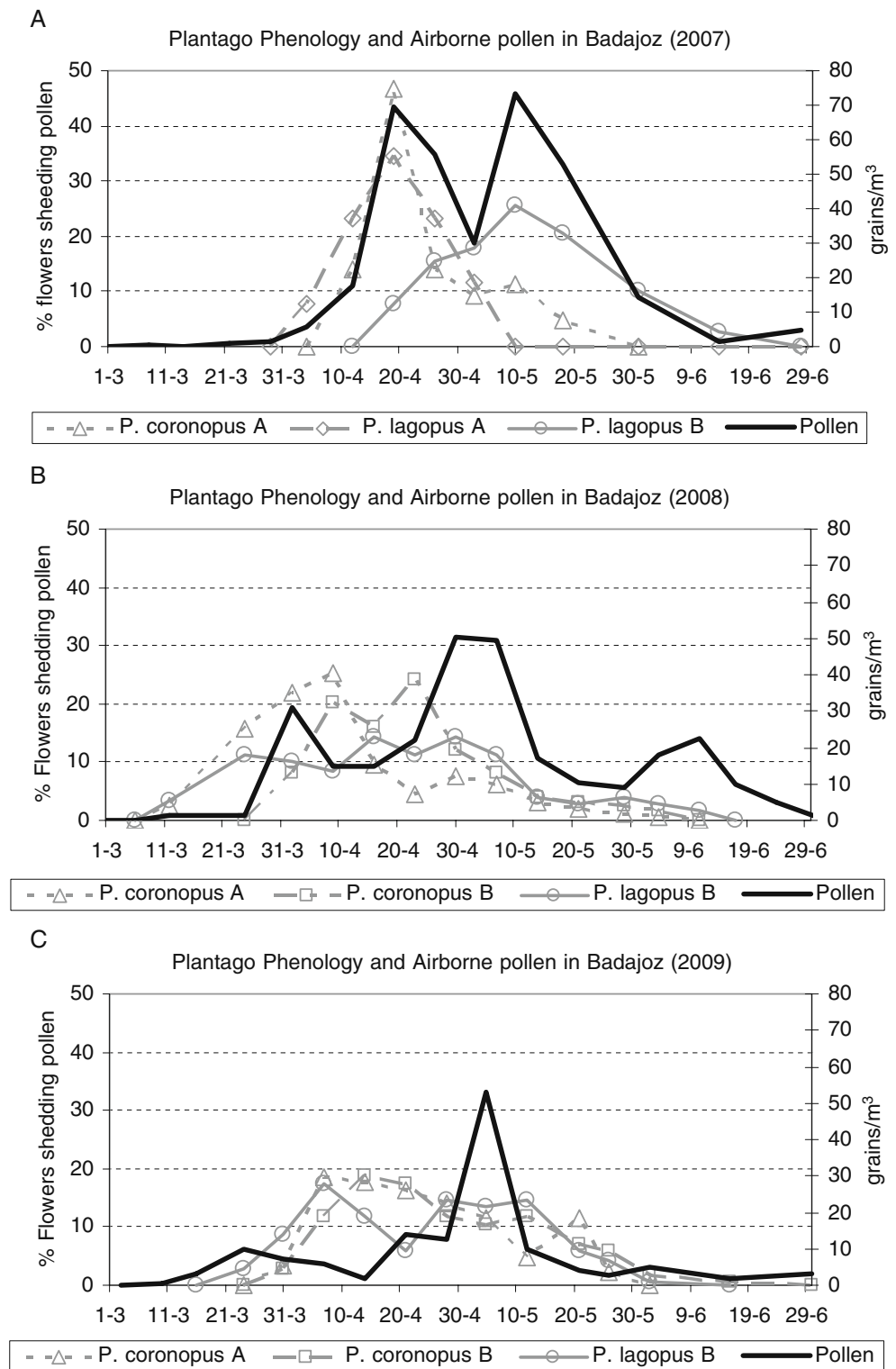


Fig. 9 Flowering phenology and airborne pollen concentration of *Plantago* species in 2007 (a), 2008 (b) and 2009 (c)



peaks and its phenology. Thus, *Cupressus sempervirens* was the main source of the captured airborne pollen. The airborne pollen grains captured outside the pollination period at the site studied probably came partly from distant sources and partly from resuspension phenomena after pollination. There is evidence of long-distance transport of Cupressaceae pollen

(Rogers and Levetin 1998; Water and Levetin 2001; Water et al. 2003) when sources are abundant in natural vegetation.

Quercus airborne pollen is known to be wind-transported far from its sources. Fairley and Batchelder (1986) found that *Quercus* pollen grains can be wind-transported for at least 16 km. Jato et al. (2002, 2007b) found that the pollen

Table 2 Poaceae species studied phenologically at the three sites (A, B, C). P Perennial, A annual

		2007			2008			2009		
		A	B	C	A	B	C	A	B	C
<i>Agrostis castellana</i>	P	*			*			*		
<i>Agrostis pourretii</i>	A	*			*					
<i>Avena barbata</i>	A	*			*	*	*	*	*	*
<i>Avena sterilis</i>	A		*	*		*	*		*	*
<i>Brachypodium distachyon</i>	A				*					
<i>Bromus diandrus</i>	A	*	*		*	*	*	*	*	*
<i>Bromus hordeaceus</i>	A	*			*	*	*	*		*
<i>Cynodon dactylon</i>	P		*	*		*	*		*	*
<i>Cynosurus echinatus</i>	A		*			*			*	
<i>Dactylis glomerata</i>	P	*			*			*		
<i>Gaudinia fragilis</i>	A	*						*		
<i>Hordeum leporinum</i>	A	*	*	*	*	*	*	*	*	*
<i>Lamarckia aurea</i>	A	*			*			*	*	*
<i>Lolium rigidum</i>	A		*	*	*	*	*	*	*	*
<i>Molineriella minuta</i>	A	*			*			*	*	
<i>Phalaris minor</i>	A	*	*	*		*	*		*	*
<i>Piptatherum miliaceum</i>	P		*	*		*	*		*	*
<i>Poa annua</i>	A			*				*		*
<i>Rostraria cristata</i>	A					*				
<i>Sorghum halepense</i>	P			*						
<i>Stipa capensis</i>	A	*			*		*	*		
<i>Trisetum paniceum</i>	A	*	*	*	*	*	*	*	*	*
<i>Vulpia geniculata</i>	A	*	*	*	*	*	*	*	*	*
<i>Vulpia myuros</i>	A	*			*			*	*	*

they captured reflected the influence of regional-scale transportation from trees in areas 10–30 km away from the sporetrap. Our estimate was that considerably more than 50% of the *Quercus* pollen type captured belonged to *Quercus rotundifolia*, mainly reflecting its representation in the vegetation (Rivas-Martínez 1987), and because of its predominance in the phenologic and airborne data. For this pollen type, the great expanse of terrain in the region with holm and cork oak could explain the presence of pollen for

several weeks after pollination in the study area, even from more than 50 km away.

The phenology of grass species is not easy to determine because of the many species involved in this group, both annuals and perennials, not all of which open their spikelets to shed pollen. Ickovic et al. (1989) studied 18 grass species over 2 years, and found that most of them flowered 2–3 weeks before any noticeable atmospheric pollen concentration. Zanotti and Puppi (2000) studied nine grass

Table 3 Spearman coefficient correlation and P-value between airborne pollen and phenological data

	2007		2008		2009	
	r	P	r	P	r	P
Cupressaceae			0.577	0.063	0.8857	0.000*
<i>Platanus</i>	0.730	0.011*	0.870	0.002*	0.849	0.002*
<i>Quercus</i>	0.742	0.002*	0.713	0.003*	0.0927	0.000*
<i>Olea</i>	0.878	0.001*	0.793	0.001*	0.617	0.011*
Poaceae A	0.381	0.132	0.213	0.381	-0.147	0.587
Poaceae B	0.624	0.007*	0.107	0.663	0.141	0.602
Poaceae C	0.477	0.053	0.055	0.825	0.112	0.680
<i>Plantago</i>	0.895	0.001*	0.619	0.006*	0.543	0.030*

*Significant at P< 0.05

species, but examined their flowering in relation to topographic gradient rather than pollen production. There are great differences in pollen production in Poaceae species (Prieto-Baena et al. 2003; Aboulaich et al. 2009). Some of these species are widespread and very abundant in vegetation, but they rarely open their spikelets, examples being most *Bromus* species, *Vulpia myuros*, and *Hordeum leporinum*. Only a few species are responsible for most of the pollen produced, with perennials having the greatest production so that their importance in airborne pollen must be taken into account; this also means that when the temperature reaches a certain threshold they do not suffer (wilt or die) at the same level as do annuals.

It is broadly accepted that the presence of pollen in the atmosphere is caused by both local and distant sources. The problem is to quantify the relative importance of each (Jato et al. 2007a). Long-distance transport may complicate the use of phenological observations in pollen forecasting, and it is known that phenological observations alone do not suffice to determine the timing of the main pollen season, at least for birch pollen (Ranta et al. 2006). It also needs to be borne in mind that both phenophase definitions and observational methods can vary from one country to another. Latorre (1999) noted that pollen data of anemophilous species coincide fairly well with phenological behaviour, although particular features must be taken into account to interpret this relationship. According to that study, while the flowering pattern is regular and constant year after year, the pollen pattern is not, especially in the final phase. Another factor is the urban effect, i.e. flowering is earlier in urbanised zones than in corresponding rural zones (Roetzer et al. 2000). This may need to be considered when the same species appears in both urban and rural contexts. In the present study, this may be the case for olive, plantain, and some grass pollen grains. The good correlation between airborne pollen and phenology data found by Hidalgo et al. (2003) for Cupressaceae and by Fornaciari et al. (2000) for some olive cultivars was probably due to the fact that there were no other pollen sources surrounding the area studied or because weather conditions did not favour long-distance transport. Apart from long-distance transport, another possible reason for the differences between the pollen and flowering data could be methodological constraints, such as not considering all the pollen sources in the phenological studies (Estrella et al. 2006). For Skjøth et al. (2007) the most effective way to improve pollen forecasts is to extend current forecasts with atmospheric transport models that take into account pollen emission and transport from neighbouring regions. This is important for allergy patients because, unless long-range transport is taken into account, pre-season pollen episodes will have a full allergic impact since the patients will, in general, be unprotected at that time. In some cases, maximum

densities of airborne pollen were recorded after the flowering period of local sources (Kasprzyk 2003). Other than long-distance transport, one must also consider resuspension effects to explain the airborne pollen recorded outside the flowering period of the local sources (Latorre 1997). This would be consistent with the observation that pollen grains always appear in the air after local sources have finished flowering, and only occasionally, and to a lesser extent, before they start. Another idea that supports the importance of resuspension is that predominant winds in the study area are from the west, not from areas where flowering has presumably already happened (south) or will happen later (north), and thus could have transported these pollen grains. Besides, more deformed pollen grains are observed at the end of pollination period than in the beginning.

Conclusions

Phenological studies have shown that airborne pollen results from both local and distant sources, although the pollen peaks usually appear when local sources are shedding the greatest amounts of pollen. The observed differences between phenology and aerobiology data can be explained by a buffer effect between the shedding of pollen and its presence in the atmosphere, by the different frequencies of phenology records (weekly) and pollen trap records (continuous), and by the different number of species involved in each pollen type. The buffer effect could be interpreted as the time delay between pollen liberation and its transportation by wind. Phenological records are a useful tool to help identify airborne pollen sources at species level, and this information could be used to improve pollen forecasts. Nevertheless, when the pollen type includes a broad range of species, it is difficult to delimit their relative importance. Phenology records would improve the usefulness of aerobiology when quantitative information on flowering is available for comparison with airborne pollen concentrations, enabling airborne pollen peaks to be evaluated at species level. Resuspension phenomena may be more important than long-distance transport in explaining airborne pollen presence outside the flowering period, especially at the end of the pollination period.

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