

## Airborne behaviour of *Echium* pollen

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### Abstract

Species of *Echium* are clearly entomophilous, but they release great amounts of pollen into the atmosphere with its consequent anemophilous transport, because their high pollen production, the smallness of the grains, and the exserted position of their anthers. Using three volumetric airborne sporetraps in Extremadura (SW of Spain) between 1994 and 1998, we found that *Echium* pollen reached pollen concentrations similar to or greater than other anemophilous plants. The main pollination period appeared from April to June. The maximum peak daily concentration reached 35.9 grains/m<sup>3</sup> and the annual recorded totals showed interannual variations between 64.2 and 614.4. Correlations were calculated between the daily pollen concentrations and the meteorological parameters rain, temperature, wind direction and velocity and relative humidity. Wind direction seems to be significant, warm dry air seems to facilitate the release of pollen into the atmosphere and increase its concentrations. Hourly pollen concentration reached a maximum between 11:00 and 12:00 and a minimum at 07:00, and the patterns were very similar in the three localities studied. This would indicate that the presence of *Echium* pollen in the atmosphere is related to the processes of anthesis of the populations near the traps, and would not correspond to a model of transport from distant zones.

### 1. Introduction

Like the rest of the species of the family *Boraginaceae*, the species of *Echium* are clearly entomophilous in the characters of their flowers and are assiduously sought out by bees and bumblebees for their high degree of nectar production (Corbet and Delfosse, 1984; Corbet et al., 1991). This makes them one of the most important contributors to the honeys of Extremadura: their *Echium* pollen content is usually around 10–15%, although it may surpass 25% (Montero and Tormo 1990, 1993).

According to Luque (1995), in Extremadura (SW Spain) there exist eight species of the genus

*Echium*: *E. boissieri* Steudel, *E. flavum* Desf., *E. plantagineum* L., *E. lusitanicum* L., *E. vulgare* L., *E. creticum* L. and *E. tuberculatum* Hoffmanns. & Link.. Nevertheless, the most abundant among them is without doubt *E. plantagineum* L., which is a major component of the nitrophiles in the pastures and meadows of the Region (Devesa and Ruiz, 1995).

Palynologically, most of these species belong to the same pollen type described by Díez (1984, 1987), characterized by being small in size and heteropolar. While this facilitates their rapid identification, it does not allow the different species to be separated aeropalynologically. The high

pollen production of these species, the smallness of the grains, and the exerted position of their anthers lead to great amounts of pollen being released into the atmosphere with its consequent anemophilous transport.

In aeropalynological studies carried out in Spanish localities, *Echium* pollen always represents less than 1% of the pollen found. The greatest value was found in Estepona in 1996, where it reached 0.96% (Cabezudo et al., 1998). With respect to the annual total concentrations of this pollen, the localities presenting the greatest concentrations are Badajoz and Mérida, followed by Cáceres, Málaga, and Estepona (Cabezudo et al., 1998; Moreno et al., 1998a, b; Recio et al., 1998; Silva et al., 1998a, b; Tavira et al., 1998a, b; Toro et al., 1998; Trigo et al., 1998).

It is worthy of note that the concentration levels of this pollen at the sites studied in Extremadura are similar to or greater than those recorded for other strictly anemophilous taxa with equal abundance. Thus, in Badajoz during the years 1993, 1994 and 1995, the *Echium* pollen levels were similar to those of *Populus* and *Scirpus*, and greater than those recorded for *Ulmus* and *Mercurialis* (Silva, 1996). This is reason to suppose that wind-borne transport of the pollen of these *Echium* species might play a relevant role in their reproduction and in genetic exchange between non-neighbouring populations. The aim of this work is attempt to know the importance of anemophily in this entomophilous genus.

## 2. Materials and methods

The study was performed using Burkard-type volumetric traps (Hirst, 1952) situated in Badajoz (38°53' N 6°58' W), Mérida (38°55' N 6°20' W), and Cáceres (39°29' N 6°22' W). These provide daily and hourly samples of the particulate concentrations in the air. The samples were analyzed by performing four longitudinal scans under optical microscopy at 400× magnification, yielding the hourly and daily pollen concentrations in grains/m<sup>3</sup>.

In Badajoz, the study was performed continuously from 1994 to 1998, whereas for the other two sites it was only performed from 1996 to 1998. There was an interruption in data collection in Mérida from 27 May to 5 June 1996 due to technical problems.

Meteorological data were provided by the Centro Meteorológico Territorial de Extremadura. These consisted of the maximum, minimum and mean temperatures (°C), relative humidity (%), rainfall (mm), wind-path (hm), and the periods of calm and of winds from the NE, SE, SW, and NW quadrants (hours). These data were collected for the observatories of Badajoz, Mérida, and Cáceres.

The study of the time of day of the appearance of *Echium* pollen in the atmosphere was carried out using the global data from the Badajoz, Mérida and Cáceres observatories in each of the years of the study, calculating for each site the mean concentration for each hour of the day, using only those in which the type was represented. The 95% confidence intervals were also calculated.

Correlations between the pollen concentrations and the meteorological parameters were studied using the data relative to the main pollination period determined as being from the day when accumulated pollen concentrations reached 5% to the day on which they reached 95% of each year's accumulated total (Nilsson and Persson, 1981).

To study which factors might affect the inter-annual differences in airborne *Echium* pollen and the differences in the phenology of their main anemophilous pollination period, we studied the correlations with the monthly means of the mean daily temperatures and with each month's rainfall from September to June for each of the periods of the study in Badajoz, as this was the location for which we had most years of data available. For these analyses the Bonferroni correction has been applied because of the high number of correlations calculated in order to avoid false positive correlations.

## 3. Results

With respect to the phenology of the airborne *Echium* pollination at the three sites of the study, Figure 1 shows the most relevant data for the annual total concentrations and the main pollination periods for the different years of the study (except for the case of Mérida 1996 in which this period could not be calculated due to the aforementioned technical problems).

There was airborne pollen transport recorded at the three locations between April and June. The onset of the main pollination period varied between 30 March in 1997 in Cáceres and 24 April

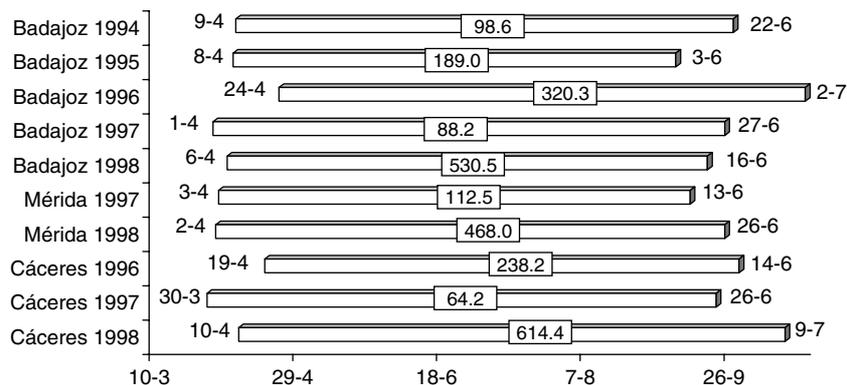


Figure 1. Total accumulated concentration of *Echium* pollen in Badajoz, Mérida, and Cáceres for each of the years studied, dates of the beginning and end of the main pollination period.

in Badajoz 1996. The end of the period varied between 3 June in Badajoz 1995 and 9 July in Cáceres 1998. The shortest main pollination period was in Badajoz 1995 with a duration of 57 days, while the longest was of 91 days in Cáceres 1998. Using Wilkerson rank test there is no significant difference ( $t = 2.803$ ,  $p = 0.0051$ ) between these main pollination periods.

With respect to the annual recorded amounts of *Echium* pollen, Figure 1 shows the existence of interannual variations for a given site. In Badajoz, the variations were between 88.2 in 1997 and 530.5 in 1998. The data obtained for Mérida were within the range of variation found for Badajoz, while in Cáceres the annual concentrations varied between 64.2 in 1997 and 614.4 in 1998.

There is no significant correlation between temperatures and rainfall with pollination. Neither meteorological factor presented any significant correlation with the date of onset of the main pollination period, although the factor that came closest to significance was the mean temperature for March ( $p = 0.0613$ ), which was negatively correlated.

The maximum values of the daily *Echium* pollen concentrations were found to be below 25 grains/m<sup>3</sup> in all the years except 1998, in which year all three sites also presented their annual concentration maxima. For this reason, the said year was chosen to compare the variations in the daily concentrations at the different locations (Figure 2). The Pearson correlation between the

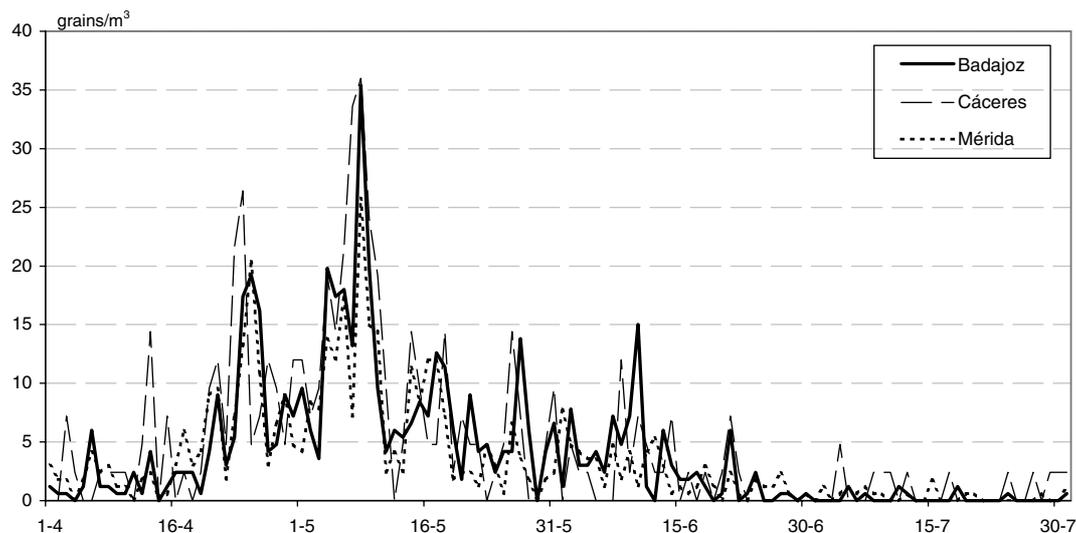


Figure 2. Variations in the daily concentrations of *Echium* pollen during 1998 for the three locations studied (grains/m<sup>3</sup>).

three locations using daily pollen concentration are significant (Badajoz–Mérida  $r = 0.842$ ; Badajoz–Cáceres  $r = 0.784$ ; Mérida–Cáceres  $r = 0.710$ ; always  $p = 0.000$ ), using the Tukey test to perform a pairwise comparison means we cannot refuse the mean similarity hypothesis (Badajoz–Mérida  $p = 0.525$ ; Badajoz–Cáceres  $p = 0.970$ ; Mérida–Cáceres  $p = 0.560$ ). One sees that there is coincidence in the magnitudes recorded at the three locations and in the dates at which the greatest concentrations were reached. Thus, in Badajoz the maximum of daily pollination was reached on 8 May with 35.9 grains/m<sup>3</sup>; in Mérida it appeared on the same day at a concentration of 26.1 grains/m<sup>3</sup>; and Cáceres, which also coincided in the date, presented 36.0 grains/m<sup>3</sup>. There also exists parallelism in the behaviour of the three observatories in the secondary peaks that appeared between 24 and 25 April, reaching concentration levels of 19.4, 20.7 and 26.4 grains/m<sup>3</sup>, respectively, as well as in other lesser peaks and in periods of decline of the pollination levels.

To determine which factors influence the daily variations, we performed a correlation analysis between the daily values of *Echium* concentrations within the main pollination period common to the three sites during 1998 (10 April–16 June) and the values of the different meteorological parameters recorded at each of the localities. The results are listed in Table 1. The only correlations found to be significant with respect to variations in daily concentrations, using the Bonferroni correction, are the periods of winds from quadrant 2 (SE) in Cáceres, the maximum temperatures in Badajoz, and negative correlations with the relative

humidity in Badajoz and Merida, although there are other high values of correlation that could evidence a general tendency in this way.

With respect to the hours of appearance of this pollen in the atmosphere, Figure 3 shows the behaviour recorded at the three sites. The pattern is independent of the site: the lowest concentrations are recorded at 07:00, after which they rise to a maximum between 11:00 and 12:00; a plateau is maintained until 18:00 or 19:00, when the levels begin to fall during the night to the minimum values at 07:00. Pearson correlation between locations using hourly data shows a strong positive association (Badajoz–Mérida  $r = 0.888$ ; Badajoz–Cáceres  $r = 0.867$ ; Mérida–Cáceres  $r = 0.803$ ; always  $p = 0.000$ ).

#### 4. Discussion and conclusions

The levels of anemophilous pollination of *Echium* in Extremadura present a high degree of parallelism with respect to their presence at the three locations studied, showing similar interannual variations and concentration levels, both as annual accumulated values and as daily values. This finding could be interpreted by supposing that, over the area of the study, the release of this pollen into the atmosphere is similar in intensity and that environmental variations affect the pollen concentrations uniformly.

The interannual variations found in the study could to be related to meteorological variations that might determine the greater or lesser development of these species. Thus, autumn rains, and in the present case in particular it seems to be the

Table 1. Study of the correlations between the values of daily *Echium* pollen concentrations in the period from 10 April to 16 June 1998 at the three locations ( $n = 68$ ), and the values of the different meteorological parameters;  $r$  is the correlation coefficient, and  $p$  the probability that  $r = 0$

	Mean T.	Max.T.	Min.T.	R.H.	Rain	Wind	Calms	NE	SE	SW	NW
Badajoz											
$r$	0.271	0.335	0.148	0.390	0.191	0.131	0.060	0.166	0.282	0.225	0.071
$p$	0.0255	<u>0.0053</u>	0.2273	<u>0.0010</u>	0.1180	0.2877	0.6288	0.1760	0.0199	0.0645	0.5633
Mérida											
$r$	0.201	0.296	0.025	0.353	0.224	0.086	0.043	0.226	0.302	0.303	0.083
$p$	0.0998	0.0142	0.8389	<u>0.0031</u>	0.0663	0.4839	0.7288	0.0635	0.0123	0.0119	0.5006
Cáceres											
$r$	0.093	0.194	0.041	0.207	0.199	0.033	0.064	0.146	0.324	0.203	0.077
$p$	0.4527	0.1131	0.7377	0.0908	0.1046	0.7885	0.6057	0.2359	<u>0.0070</u>	0.0975	0.5328

Significant correlations using the Bonferroni correction are underlined.

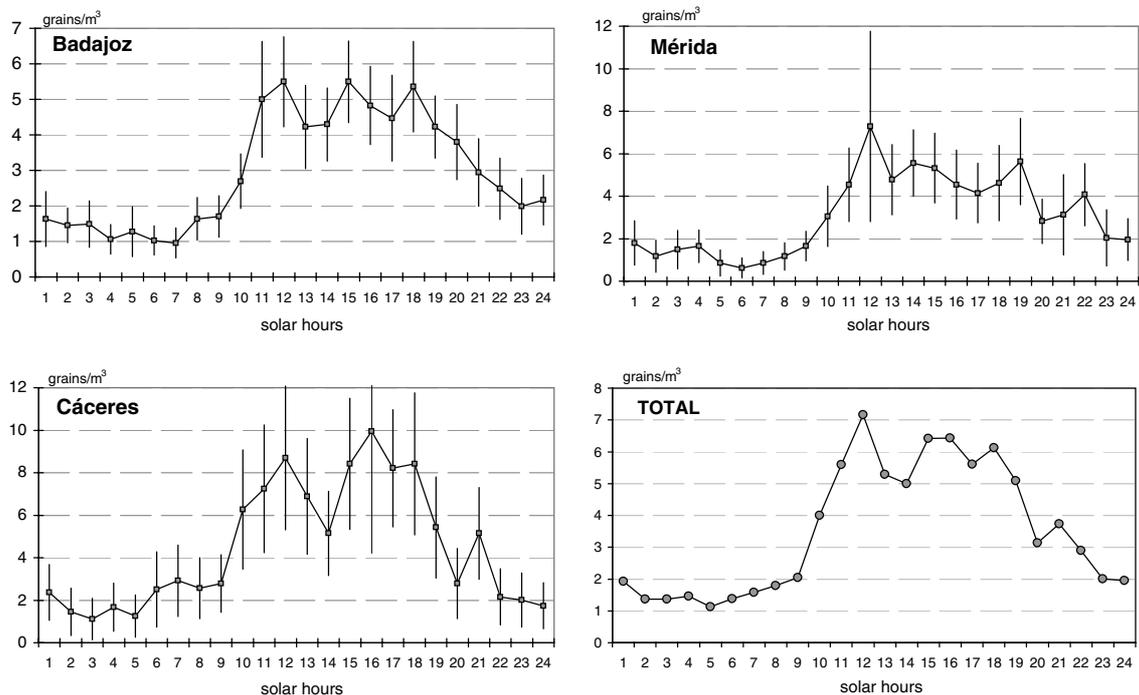


Figure 3. Hourly variations in the concentrations of *Echium* pollen for the three locations studied, as well as for their sum. The means and 95% confidence intervals are shown for the values reached on those days on which the pollen type was observed.

November rains, would favour the development of these plants, especially of annual species such as *E. plantagineum* which is the most abundant in the Region. This finding has been reported for many other taxa, in particular for the levels of annual pollination of various herbaceous species in Extremadura (Silva et al., 1998).

Intraannually, the daily variations in *Echium* concentrations are related in the three locations with the presence of winds from the SE quadrant. In principle, this finding might indicate a contribution of this pollen from zones located in the said sector. However, given the abundance with which at least *E. plantagineum* is present throughout the Region, it is difficult to see that the cause of the correlation would be pollen imports. Since there are positive correlations in Badajoz and Mérida with the maximum temperatures and negative correlations with the relative humidity, and given that the three sites always present positive correlations with winds from the E and negative correlations with winds from the W, it is easier to suppose that it is in reality warm dry air which facilitates the release of pollen into the atmosphere and increases its concentrations. This correlation

appears frequently in pollination processes studied by other workers, and is positive in the Peninsula in pollinations that take place before the month of July when the temperatures in the Mediterranean Region begin to be extreme (Martín et al., 1990; Fernández et al., 1993). The case of Extremadura is accentuated when the winds are from the interior of the Peninsula, as against winds from the W which are usually more moisture laden.

That the three locations present such a marked and uniform pattern of hourly variation would also indicate that the presence of *Echium* pollen in the atmosphere is related to the processes of anthesis of the populations near the traps, and would not correspond to a model of transport from distant zones (Trigo et al., 1997). The model that is presented corresponds to a taxon with diurnal anemophilous pollination, coinciding with the model of entomophilous pollination of *Echium* species.

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## References

- Cabezudo B., Toro F.J., Recio M. and Trigo M.M.: 1998, Aerobiología en Andalucía: Estación de Estepona (1995–1996). *REA* **3**, 37–40.
- Corbet S.A. and Delfosse E.S.: 1984, Honeybees and the nectar of *Echium plantagineum* L. in the south-eastern Australia. *Aust. J. Ecol.* **9**, 125–139.
- Corbet S.A., Williams I.H. and Osborne J.L.: 1991, Bees and the pollination of crops and wild flowers in the European Community. *Bee World* **72**, 47–59.
- Devesa J.A. and Ruiz T.: 1995, Vegetación, in J.A. Devesa (ed), *Flora y Vegetación de Extremadura*. Universitas Editorial: Badajoz, pp. 81–115.
- Diez M.J.: 1984, Contribución al Atlas palinológico de Andalucía Occidental. I. *Boraginaceae*. *Lagascalia* **13**, 147–171.
- Diez M.J.: 1987, Boraginaceae, in B. Valdés, M.J. Diez, and I. Fernández, (eds) *Atlas polínico de Andalucía Occidental*. Instituto de Desarrollo Regional de Universidad de Sevilla. Utrera, pp. 265–281.
- Fernández D., Suárez M., Díaz T. and Valencia R.M.: 1993, Airborne pollen and spores of León (Spain). *Int. J. Biometeorol.* **37**, 89–95.
- Hirst J.M.: 1952, An automatic volumetric spore trap. *Ann. Appl. Biol.* **39**, 257–265.
- Luque T.: 1995, *Echium*, in J.A. Devesa (ed), *Flora y Vegetación de Extremadura*. Universitas Editorial. Badajoz, pp. 450–451.
- Martín J., Suárez-Cervera M. and Seoane-Camba J.A.: 1990, Influencia de los factores climáticos en la captura del polen aerovagante. In G. Blanca and C. Díaz de la Guardia (eds), *VII Simposio de Palinología Polen, Esporas y sus Aplicaciones*, Granada, pp. 385–390.
- Montero I. and Tormo R.: 1990, Análisis polínico de mieles de cuatro zonas de montaña de Extremadura. *An. Asoc. Palinol. Leng. Española* **5**, 71–78.
- Montero I. and Tormo R.: 1993, Preferencias polínicas de la abeja en un colmenar en el Sur de Badajoz. *An. Asoc. Palinol. Leng. Española* **6**, 93–102.
- Moreno A., Muñoz A.F., Tormo R. and Silva I.: 1998a, Aerobiología en Extremadura: Estación de Mérida (1996). *REA* **3**, 65–68.
- Moreno A., Muñoz A.F., Tormo R. and Silva I.: 1998b, Aerobiología en Extremadura: Estación de Mérida (1997). *REA* **4**, 99–102.
- Nilsson S. and Persson S.: 1981, Tree pollen spectra in the Stockholm region (Sweden), 1973–1980. *Grana* **20**, 179–182.
- Recio M., Trigo M.M., Toro F.J. and Cabezudo B.: 1998, Aerobiología en Andalucía: Estación de Málaga (1997). *REA* **4**, 41–44.
- Silva I.: 1996, *Estudio aeropalinológico de la ciudad de Badajoz*. Tesis Doctoral. Universidad de Extremadura.
- Silva I., Muñoz A.F. and Tormo R.: 1998a, Aerobiología en Extremadura: Estación de Badajoz (1997). *REA* **4**, 95–98.
- Silva I., Muñoz A.F., Tormo R. and Recio D.: 1998b, Aerobiología en Extremadura: Estación de Badajoz (1995–1996). *REA* **3**, 61–64.
- Silva I., Tormo R. and Muñoz, A.F.: 1998c, Differences in phenology and pollination levels between dry and wet years. Badajoz (SW Spain). *6th International Congress on aerobiology*. Perugia, pp. 292.
- Tavira J., Tormo R., Silva I. and Muñoz A.F.: 1998a, Aerobiología en Extremadura: Estación de Cáceres (1996). *REA* **3**, 69–72.
- Tavira J., Tormo R., Silva I. and Muñoz A.F.: 1998b, Aerobiología en Extremadura: Estación de Cáceres (1997). *REA* **4**, 103–106.
- Toro F.J., Recio M., Cabezudo B. and Trigo M.M.: 1998, Aerobiología en Andalucía: Estación de Estepona (1997). *REA* **4**, 45–48.
- Trigo M.M., Recio M., Toro F.J. and Cabezudo B.: 1997, Intradurnal fluctuations in airborne pollen in Málaga (S. Spain): A quantitative method. *Grana* **36**, 39–43.
- Trigo M.M., Recio M., Toro F.J. and Cabezudo B.: 1998, Aerobiología en Andalucía: Estación de Málaga (1995–1996). *REA* **3**, 33–36.