

Changes in the Mediterranean pine forest: pollination patterns and annual trends of airborne pollen

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Abstract In some areas, forests are being affected in diverse aspects such as structure, composition and biodiversity showing an increase or a decrease in the growth rates. *Pinus* is one of the most dominant genera in the forests of the Northern Hemisphere. This study analyzes the pine pollination patterns in 30 locations of Spain with an average of 21-year dataset. The aim is to evaluate possible changes in flowering intensity as well as in annual pollen production trends, according to the airborne pollen patterns. Annual Pollen Indices

show three threshold values: (1) over 4000 grains per year in Catalonia, the Central System Mountains and Ourense (Galicia), (2) between 4000 and 1000 grains in central-south Spain and in the Balearic Islands, and (3) under 1000 in eastern Spain, Cartagena and the Canary Islands. Airborne pollen patterns were also influenced by *Pinus* species: The species located in the littoral and low land areas pollinated in the first pollination phase, from February to April, and the mountain pine species did in the second one, from

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April to June. The statistical analyses reveal increasing significant trends in 12 sites and significant decreasing trends in two. The *Pinus* flowering intensity is showing an earlier start and a delay in the end of the pollination period, thus a longer period of pollen in the air. This study suggests that the aerobiological monitoring is an interesting bio-indicator of changes happening in *Pinus* landscapes, and therefore explains the vulnerability of this genus in Spain.

Keywords *Pinus* pollen · Environment and climate change · Flowering intensity · Annual trends · Pollination patterns

1 Introduction

Recent studies on the effect of climate change on forest ecological conditions have demonstrated that in some areas the forest growth rates are increasing although in others they decrease (FAO 2011). Europe was the only worldwide region with an evident increase of the surface of forested area over the period 1990–2010, due in large part to a few countries, led by Spain (118,500 ha per year) (FAO 2011). However, several studies have recognized that the climate change affects directly and indirectly the forest through disturbances (Dale et al. 2000) which act independently or in combination with others factors as the atmospheric changes in chemical composition, or changes by fires or disease outbreaks (Lindner et al. 2010). Moreover, in addition to the natural disturbances, the human activities show a globally significant impact on biological systems (Rosenzweig et al.

2008). The analysis of the mountain vegetation alterations should be made with care because there are several factors that confuse the climate signal, such as the species interactions, the dynamic nature of biodiversity, the dispersal capacity of tree species, the abandonment of silviculture or the habitat modification (Lenoir et al. 2010).

Pinus is a dominant genus of the Northern Hemisphere forest, promoted for the natural distribution and silvicultural importance. In Spain, *Pinus* is mostly represented by eight species distributed from the littoral and low land areas (*P. canariensis* Chr. Sm. ex DC., *P. halepensis* Mill., *P. pinaster* Aiton, *P. pinea* L. and *P. radiata* D. Don) to the mountain areas (*P. nigra* J.F. Arnold, *P. sylvestris* L.) and subalpine regions (*P. uncinata* Ram.). All these species are trees which produce anemophilous pollen of large size constituted by a central body and two laterally placed vesicles or sacs that assist the atmospheric flotation and its dispersion.

Several Spanish studies showed that *Pinus* pollen is one of the most abundant airborne pollen taxa during the seasons of spring and summer (Ruiz Garcia et al. 1998; Rodríguez-Rajo et al. 2003; Docampo et al. 2007; Pérez Badia et al. 2010). Monitoring and measuring the airborne pollen is a multidisciplinary tool which provide information on plant phenology as well as its distribution (Galán et al. 2016), with application in the potential crop production, the health care and the biodiversity conservation (Belmonte et al. 2008; Tormo-Molina et al. 2011; Fernández-González et al. 2011; Fernández-Llamazares et al. 2014). As regards the conservation, airborne pollen is used to improve the bio-geographical and ecological information of plants, being important for the protection of the

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plant biodiversity (Womack et al. 2010; Cariñanos et al. 2010; Alba-Sánchez et al. 2010), as well as it is a sensitive indicator of climate change (Clot 2003; García-Mozo et al. 2010; Cariñanos et al. 2013; Fernández-Llamazares et al. 2014). The inter-annual pollination trends usually showed a relationship with the climate variability (Cariñanos et al. 2010; Fernández-Llamazares et al. 2014); therefore, airborne pollen could be a powerful tool to study environmental changes, especially the vegetation distribution. Some authors also pointed out that the influence of climate change could be evaluated according to airborne pollen data from trees or herb plants, i.e., in Europe (Green et al. 2003; Clot 2003; Cariñanos et al. 2004; Frei and Gassner 2008; Tormo-Molina et al. 2010; Ziello et al. 2012; Fernández-Llamazares et al. 2014).

The present study analyzes the Mediterranean pine pollination patterns in 30 locations of Spain with an average of 21-year dataset to determine whether pine forests are affected by environmental changes. The aim is to evaluate possible changes in flowering intensity and annual trends of pollen production according to airborne pollen patterns.

2 Materials and methods

2.1 Area of study

Pinus pollen data from 30 aerobiological stations located in different Spanish areas are analyzed (Table 1; Fig. 1a). The stations are representative of the bio-geographical diversity of Spain, representing latitudinal and regional perspectives, different climatic, topographic and vegetation characteristics.

Ourense and Santiago de Compostela are sited in Galicia region, NW Iberian Peninsula, situated in the boundary between the Euro-Siberian and Mediterranean regions. While Santiago de Compostela is located in the Euro-Siberian region with a mean annual temperature around 14.2 °C and annual rainfall of 1896 mm (data calculated for period under study, Table 1); Ourense receives the Mediterranean influence, with oscillations in temperature (summer–winter and day–night) and lower annual rainfall (15.1 °C, 835 mm). The forest cover in Galicia represents a 47.5% of the total region, and a 17.1% (506,000 ha) is composed by coniferous. The main *Pinus* species are *P. pinaster* and *P. radiata* (DGCN, IFN3 2007).

The Castilla y León region is located in the Northwest of Iberian Peninsula, and it is mainly considered as Mediterranean region. In our study, the aerobiological stations of León, Burgos, Miranda de Ebro, Salamanca, Segovia, Soria and Valladolid were analyzed. They present yearly a mean annual temperature ranging from 10.9 °C (Burgos) to 12.5 °C (Salamanca). The annual rainfall varies between 354 mm (Salamanca) to 530 mm (Burgos). Other aerobiological station of this region is Arenas de San Pedro. It is placed in the Central System Mountains and shows the warmest and rainiest conditions (15.7 °C and 761 mm). The total forested area in Castilla y León represents the 31.6% of the territory, being the coniferous forests cover by 906,000 ha (9.6% of this area) (DGCN, IFN3 2007). The native conifers are *P. pinaster*, *P. pinea*, *P. nigra*, *P. sylvestris* and silvicultured species such as *P. radiata*. (DGCN, IFN3 2007).

Eight sites studied are located in Catalonia region, in the North East of the Iberian Peninsula (Barcelona, Bellaterra, Girona, Lleida, Manresa, Tarragona, Roquetes-Tortosa and Vielha). Although the climate in Catalonia is mostly Mediterranean, some areas show continental Atlantic or even Euro-Siberian influences. Vielha, placed in a Pyrenean valley, shows the coldest and rainiest conditions (9.6 °C and 921 mm). Bellaterra, Manresa and Girona present a mean temperature around 15 °C and rainfall ranging from 554 to 670 mm. Barcelona, Tarragona and Roquetes-Tortosa share a influence by the proximity to the sea (15.5–18.1 °C and 517–596 mm). Lleida, situated inland, shows the most extreme temperatures, although mean values (14.4 °C) do not reflect it, and the lowest rainfall (318 mm). The total forested area in Catalonia represents the 51% of the territory, and 44% of this area (716,000 ha) corresponds to coniferous forests (DGCN, IFN3 2007). The dominant fraction of the Catalan pine forest is integrated by native species (*P. pinea*, *P. halepensis*, *P. pinaster*, *P. nigra*, *P. sylvestris* and *P. uncinata*), which are sometimes silvicultured or naturalized, as well as the introduced *P. radiata* (Thuiller et al. 2003).

At the central-south of the Iberian Peninsula are located the regions of Madrid and Castilla-La Mancha (Toledo). Madrid (16.7 °C and 420 mm) situated at certain altitude (667 m.a.s.l.) shows for the study period, a hotter climate than Toledo (16.2 °C and 356 mm) that present a warmer climate. The forest

Table 1 Geographical and climatic characteristics and *Pinus* species present in the aerobiological stations

Aerobiological stations (period under study)	Geographical characteristics		Climatic characteristics (Allué 1990)		Climatic characteristics (period under study)		<i>Pinus</i> spp. ¹
	Altitude (m.a.s.l)	Geographical Coordinates	Mean annual temperature (°C)	Annual Precipitation (mm)	Mean annual temperature (°C)	Annual Precipitation (mm)	
Almería (2004–2010)	23	36°50'N.02°27'W.	17.9	231	19.1	208	b, g
Arenas de San Pedro (2008–2014)	510	40°12'N.05°05'W.	14.5	1483	15.7	761	d, e, g
Badajoz (1994–2014)	186	38°53'N.06°58'W.	16.7	474	17.2	448	c, e
Barcelona (1994–2014)	93	41°23'N.02°09'E.	16.4	593	15.5	596	b, d, e, g
Bellaterra (1994–2014)	245	41°30'N.02°06'E.	15.2	594	15.4	554	b, d, e, g
Burgos (2007–2014)	929	42°20'N.03°41'W.	10.5	564	10.9	530	c, d, f, g
Cartagena (1994–2014)	627	37°36'N.00°59'W.	17.3	287	18.8	244	b, d, e
Córdoba (1994–2014)	123	37°50'N.04°45'W.	17.9	674	18.4	622	b, d, e
Girona (1996–2014)	98	41°59'N.02°49'E.	15.0	740	15.4	670	b, d, e, f, g
Granada (1994–2014)	685	37°11'N.03°35'W.	15.6	448	15.3	375	b, c, g
Izaña (2005–2014)	2367	28°18'N.16°30'W.	9.4	464	10.6	405	a, f
Jaén (1994–2014)	550	37°46'N.3°47'W.	17.0	601	17.0	453	b, d, e
León (1994–2014)	837	42°36'N.05°33'W.	9.8	965	11.2	522	c, d, g
Lleida (1996–2014)	202	41°37'N.00°35'E.	15.1	385	14.4	318	b, c, g
Madrid (1994–2014)	667	40°24'N.03°40'W.	14.4	469	16.7	420	b, d, e
Málaga (1994–2014)	6	36°43'N.04°25'W.	18.5	469	18.9	545	b, d, e, f
Manresa (1996–2014)	291	41°43'N.01°50'E.	13.6	618	15.0	562	b, c, e, g
Miranda de Ebro (2007–2014)	471	42°40'N.02°56'W.	12.0	498	11.9	634	c, d, f, g
Ourense (1994–2014)	124	42°00'N.07°51'W.	13.8	802	15.1	835	d, f, g
Palma de Mallorca (2004–2011, 2014)	32	39°57'N.02°65'E.	16.8	451	18.3	486	b
Roquetes-Tortosa (2005–2014)	14	40°49'N.00°29'E.	16.8	576	18.1	517	b, d, e
Salamanca (2007–2014)	797	40°57'N.05°39'W.	12.0	421	12.5	354	d, g
Santiago de Compostela(1994–2014)	260	42°53'N.08°32'W.	12.8	1545	14.2	1896	d, f
Segovia (2007–2014)	1002	40°56'N.04°07'W.	11.4	468	11.9	398	d, e, g
Soria (2007–2014)	1063	41°45'N.02°28'W.	10.5	572	11.1	483	c, d, g
Sta. Cruz de Tenerife (2005–2014)	37	28°28'N.16°15'W.	20.8	251	21.7	386	a, f
Tarragona (1996–2014)	44	41°07'N.01°14'E.	15.8	478	15.8	560	b, c, d, e, g
Toledo (2005–2014)	450	39°51'N.04°02'W.	15.0	375	16.2	356	b, d, e
Valladolid (2007–2014)	734	41°38'N.04°44'W.	12.0	373	11.7	405	b, d, e

Table 1 continued

Aerobiological stations (period under study)	Geographical characteristics		Climatic characteristics (Allué 1990)		Climatic characteristics (period under study)		<i>Pinus</i> spp. ¹
	Altitude (m.a.s.l)	Geographical Coordinates	Mean annual temperature (°C)	Annual Precipitation (mm)	Mean annual temperature (°C)	Annual Precipitation (mm)	
Vielha (2004–2014)	974	42°42'N.00°47'E.	9.9	899	9.6	921	g, h

¹ *Pinus* spp.: a—*P. canariensis*, b—*P. halepensis*, c—*P. nigra*, d—*P. pinaster*, e—*P. pinea*, f—*P. radiata*, g—*P. sylvestris*, h—*P. uncinata*

cover in both regions is similar (34% of the total surface approximately); in Madrid, the coniferous surface reaches the 9% (80,000 ha) and in Castilla-La Mancha a 14% (110,400 ha). However, the forest cover of Toledo province reaches 25% and the coniferous surface a 3% (44,200 ha) of the total. In both cases, the pine tree is represented by *P. halepensis*, *P. pinea* and *P. pinaster*. Also, in Madrid is presented *P. sylvestris* (DGCN, IFN3 2007).

Badajoz (Extremadura, SW Spain), located by the River Guadiana, close of the Spanish-Portuguese border, presents 17.2 °C and 448 mm. The forest cover in Extremadura is above 46% of the surface, and 6% of it corresponds to *P. pinaster*, *P. pinea*, *P. halepensis*, *P. sylvestris* and *P. radiata*, most of them silvicultured. In Badajoz the area covered by conifers is 35,000 ha (DGCN, IFN3 2007).

Palma de Mallorca (Balearic Islands) is the easternmost aerobiological station in this study. The annual mean temperature is 18.3 °C, and the annual precipitation of 486 mm. The total forested area represents the 37% of the territory, and the 18% of this area (92,000 ha) corresponds to coniferous forests of *P. halepensis* and with a very low percentage of *P. pinea* silvicultivated (DGCN, IFN3 2007).

Five Andalusian sampling sites (SE Spain) are studied. In the inland are situated Córdoba, Granada and Jaén that present greater oscillations in temperature (summer–winter and day–night). The mean annual temperature registers a ranging from 15.3 °C of Granada to 18.4 °C of Córdoba. The annual rainfall varies between 622 mm (Córdoba) to 375 mm (Granada). In the littoral are located Málaga (18.9 °C, 575 mm) and Almería (19.1 °C, 208 mm), and this last city shows the warmest and driest climates of Andalusia region. The forest cover in this region represents the 34% of the total area, and 9% of it

(781,000 ha) are coniferous forests. In the studied provinces, the coniferous forests represent 626000 ha. The more representative *Pinus* species are *P. halepensis*, *P. pinaster* and *P. pinea*; in Granada and Almería, which receive the influence of Sierra Nevada Mountain, the species of *P. nigra*, and *P. sylvestris* are present as native or servicultured species (DGCN, IFN3 2007).

Cartagena (Murcia region) is located in the South-east of the Iberian Peninsula. Together with Almería, this locality shows the warmest and driest climate of the peninsula (18.8 °C, 244 mm). The forest cover of Murcia represents 28% of the total area, and a 24% of it corresponds to *P. halepensis*, and a lower percentage of *P. pinaster* and *P. pinea* (DGCN, IFN3 2007).

The Canary Islands are the southernmost and westernmost Spanish territory located in the Atlantic sea. Two sites from the Tenerife Island (Santa Cruz de Tenerife with 21.7 °C and 386 mm, and Izaña, on the Teide Mountain with 10.6 °C, 405 mm) were analyzed. In Tenerife, the forest cover represents the 33% of the island total area, and 61% of it corresponds to conifers (68,000 ha) (DGCN, IFN3 2007). The main species are the native *P. canariensis* used also in reforestations, as well as *P. radiata* (Gobierno de Canarias 2007).

2.2 Aerobiological data

Daily samples were recorded by means of 7-day recording Hirst-type volumetric traps (Hirst 1952). The traps were working uninterruptedly, sucking air at a rate of 10 L/min. The samples obtained were analyzed following the standard methodology established by the Spanish Aerobiology Network (Galán et al. 2007) and the minimum requirements of the European Aerobiology Society (Galán et al. 2014).

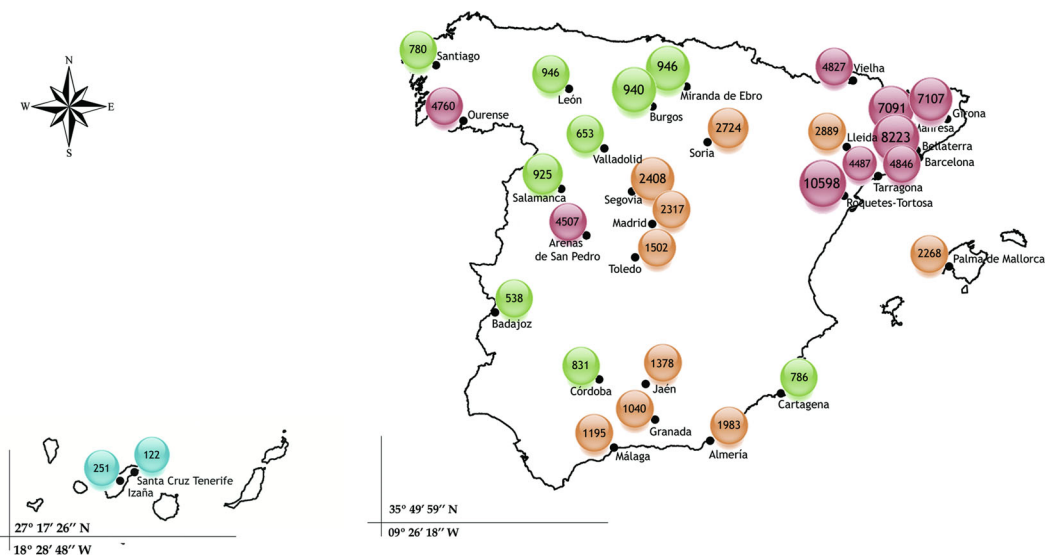
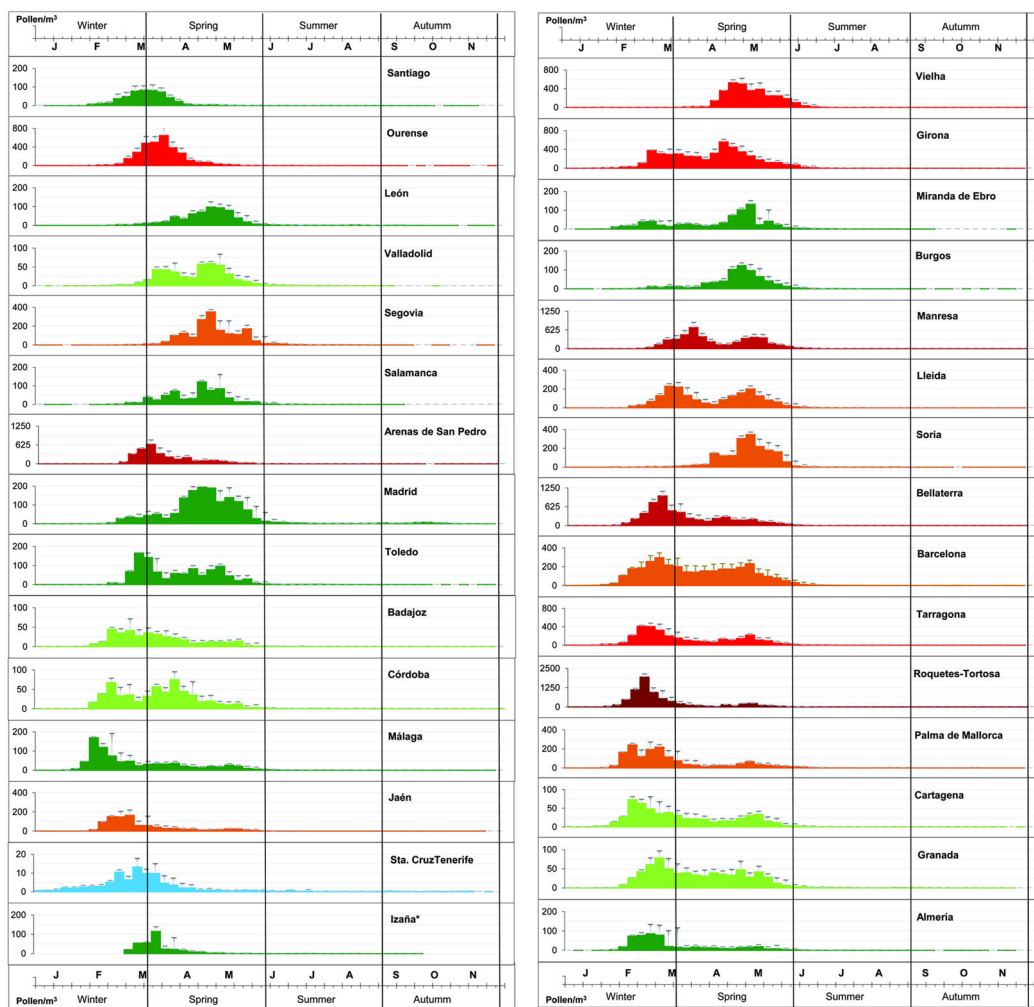
a**b**

Fig. 1 **a** Geographical situation and Annual Pollen Indices of the aerobiological stations within the study territory. **b** Yearly dynamics of the mean weekly *Pinus* pollen concentrations in the sampling stations. *First column* corresponds to eastern stations; *second column* to western stations. The *graphic colors* correspond to pollen concentration of *Y* axis. *Errors bars* have been calculated by standard deviation of each station. *Number sign* the study period is from April to October

The results were expressed as daily mean pollen concentration per cubic meter of air (pollen/m³). Annual Pollen Index (API), defined as the sum of the mean daily pollen concentrations in the year and expressed in number of pollen grains, have been calculated. In the case of Izaña (Canary Islands), a locality at an altitude of 2367 m.a.s.l., and where the sampler is only functional from April to October, we have used API for simplifying the text but when done we indicate the lengths of the sampling. We have also calculated the Monthly Pollen Index (MPI) defined as the sum of the mean daily pollen concentrations in the month and expressed in number of pollen grains. Finally, Absolute daily maximum concentration data, defined as the day with maximum pollen concentration data during study period, have been included.

The stations provided different number of yearly periods comprised between 1994 and 2014 (Table 1), depending on the available data: 21-year series for Badajoz, Barcelona, Bellaterra, Cartagena, Córdoba, Granada, Jaén, León, Madrid, Málaga, Ourense, and Santiago de Compostela; 19 for Girona, Manresa, Lleida and Tarragona; 11 for Vielha; 10 for Toledo and Santa Cruz de Tenerife; 9 for Izaña, Palma de Mallorca and Roquetes-Tortosa; 8 for Burgos, Miranda de Ebro, Salamanca, Segovia, Soria, and Valladolid; and 7 for Almería and Arenas de San Pedro.

2.3 Meteorological data

The possible influence of meteorological parameters on *Pinus* pollination was examined in each station studied (Table 1). We collected meteorological information from Phytoclimatic Atlas of Spain (Allué 1990) and also analyzed the yearly periods comprised between 1994 and 2014, depending on the aerobiological available data. Annual series of temperature (maximum, minimum and mean temperature), and total precipitation were used. Data were provided by the Agencia Estatal de Meteorología (AEMET), except in Catalonia where data were provided by

AEMET and Servei Meteorològic de Catalunya (MeteoCat). In the statistical analysis, meteorological parameters have been considered according the climate seasons of winter, spring, summer and autumn.

2.4 Statistical analyses

We have analyzed API data to evaluate the normality over the period studied by applying the Shapiro–Wilk Normality Test (SW, Shapiro and Wilk 1965). The null hypothesis is H_0 : “there is normality” against H_1 : “there is not normality.” Because some data showed non-normality (p value $<0.05^*$; $<0.01^{**}$), in order to look for the trends, we applied a combination of parametric (when normality is not refused by using SW) and nonparametric tests.

The rank-based Spearman’s Rho test, (SR, Spearman 1904) was applied in all cases by calculating the exact Rho statistic (ρ), and comparing it with the critical values (Zar 1972). In the cases where API showed normal distribution, Pearson correlation test (PC, Pearson 1896) was also calculated. These tests allowed carrying on the statistical test to contrast the null hypothesis H_0 : “there is no monotonic trend” against the (one-sided) alternative H_1 : “there is an increasing trend” or H_1 : “there is a decreasing trend,” as appropriate. As usual, significance levels at 5 and 1%, are labeled * and **, respectively.

When the SR test indicated a statistically significant monotonic trend, either increasing or decreasing, the magnitude of the trend (expressed in number of pollen grains) was estimated by using the nonparametric Theil–Sen estimator (Theil 1950; Sen 1968).

SR test was also applied to detect significant trends in *Pinus* MPI of the study period and to analyze the correlation between *Pinus* API and the meteorological data. The statistical procedures have been carried out with the help of the free software environment for statistical computing and graphics R version 2.15.0 (R Development Core Team 2007) and using an *ad hoc* spreadsheet.

3 Results

3.1 Aerobiological study

The main parameters of airborne *Pinus* pollen are shown in Table 2 and Figs. 1a and 2. An important

Table 2 Main aerobiological parameters for airborne *Pinus* pollen in each station

Aerobiological stations (years under study)	Airborne <i>Pinus</i> pollen			
	Mean Annual Pollen Index (API)		Absolute daily maximum concentration	
	Pollen grains	Pollen/m ³	Date	% <i>Pinus</i> pollen
Almería (7)	1882	659	25-Mar-05	20.0
Arenas de San Pedro (7)	4507	1044	6-Apr-08	13.0
Badajoz (21)	538	305	8-Mar-98	27.2
Barcelona (21)	4846	798	21-Apr-97	11.0
Bellaterra (21)	8223	1614	27-Mar-11	10.2
Burgos (8)	941	216	1-Jun-12	15.3
Cartagena (21)	786	261	9-Mar-98	15.7
Córdoba (21)	831	281	3-May-14	13.5
Girona (19)	7107	697	22-Mar-02	12.6
Granada (21)	1040	233	16-May-14	10.5
Izaña ^a (9)	251	356	18-Apr-14	37.8
Jaén (21)	1378	1104	25-Mar-05	38.7
León (21)	946	169	17-May-14	7.0
Lleida (19)	2889	544	5-Apr-12	9.1
Madrid (21)	2317	480	14-Jun-98	6.2
Málaga (21)	1195	1027	26-Feb-97	24.2
Manresa (19)	7091	1030	9-Apr-14	9.2
Miranda de Ebro (8)	946	293	5-Jun-14	26.3
Ourense (21)	4760	1003	25-Mar-02	15.4
Palma de Mallorca (9)	2268	710	26-Mar-10	25.3
Roquetes-Tortosa (9)	10,598	2229	9-Mar-09	15.4
Salamanca (8)	1019	497	19-May-14	13.9
Santiago de Compostela(21)	780	233	3-Mar-88	11.5
Segovia (8)	2408	860	25-May-11	22.3
Soria (8)	2724	435	20-Jun-10	10.9
Sta. Cruz de Tenerife (10)	122	32	1-Apr-14	14.1
Tarragona (19)	4487	1098	16-Mar-14	10.8
Toledo (10)	1502	503	1-Apr-14	18.7
Valladolid (8)	653	131	20-May-14	11.0
Vielha (11)	4827	701	23-May-12	8.3

^a The study period is from May to October

difference between the regions analyzed was observed. The northeastern stations (Catalonia) registered API values over 4000 pollen grains (except in Lleida that presented 2451 pollen grains), as well as Arenas de San Pedro (in the Central System Mountain) and Ourense (north-western Spain) with 4507 and 4760 pollen grains, respectively. The center and southern Peninsula and the Balearic Islands API varied between 2734 pollen grains in Soria and 1040

in Granada. In the western localities, values were lower than 1000 pollen grains. Finally, both stations of the Canary Islands registered the lowest API values, under 300 pollen grains.

Absolute daily maximum concentration occurred in different dates between February and June. The earliest peak date was registered in Málaga (February 26, 1997) and the latest in Soria (June 20, 2010). The percentage of pollen registered in the day of the

Fig. 2 Boxplots of the *Pinus* Annual Pollen Index. Stations are distributed by median value. Red boxplot Median of *Pinus* pollen ≥ 4000 ; Orange boxplot $1.000 \leq$ *Pinus* pollen < 4000 ; Green boxplot *Pinus* pollen < 1.000

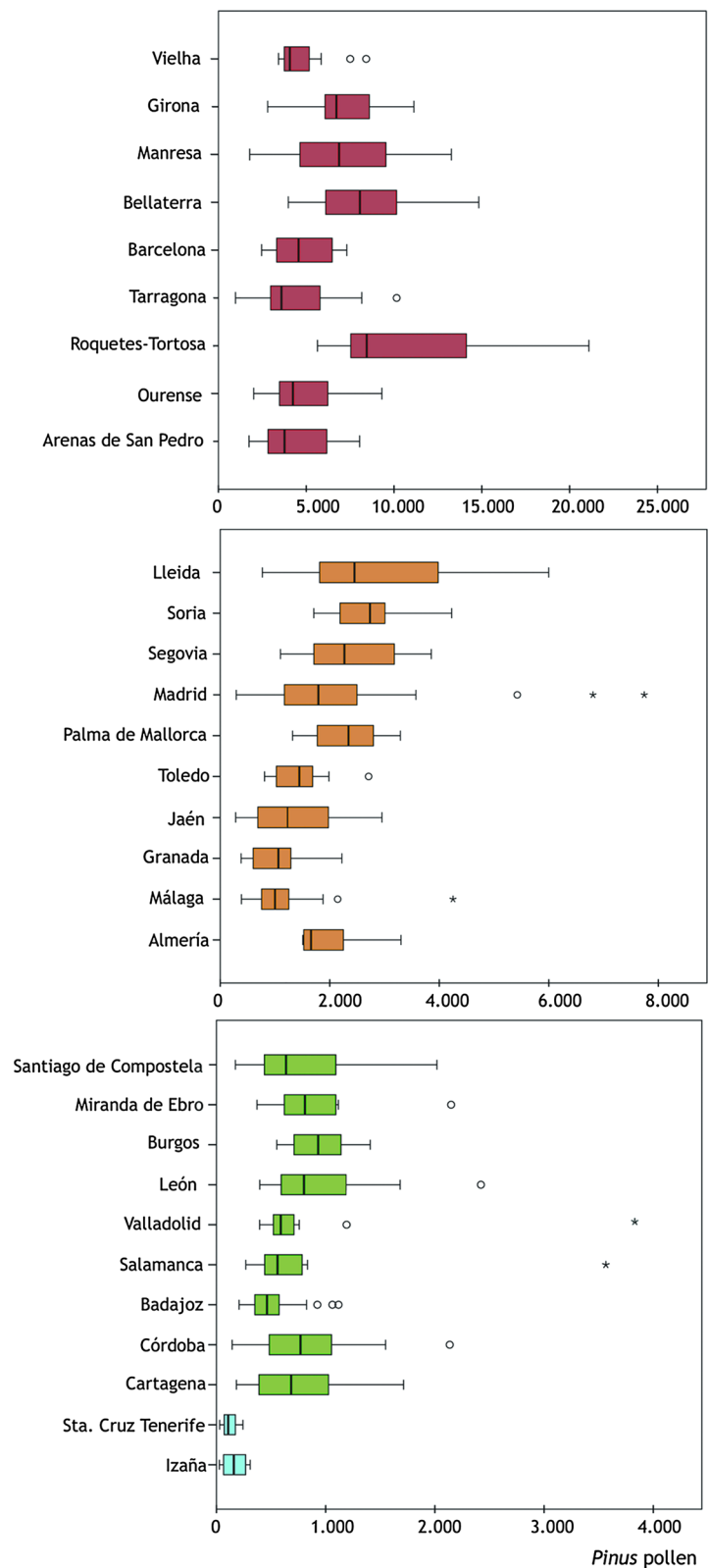


Table 3 Trends of the *Pinus* Annual Pollen Indices (API)

	Shapiro–Wilk Normality test ¹	Pearson' correlation test ²			Spearman's Rho test ³			Theil–Sen estimator ⁴		
Aerobiological station (years under study)	p-value	r	p-value	Result	ρ statistic	Critical values C1			Variation in the number of pollen grains per year	Pollen index variation (%)
						5%	1%	Result		
Badajoz (21)	0.020*				−0.682	0.370	0.508	Decreasing**	−24	−4
Barcelona (21)	0.049*				0.197			NS		
Bellaterra (21)	0.518	0.440	0.046	Increasing*	0.384			Increasing*	209	3
Cartagena (21)	0.029*				0.515			Increasing**	35	4
Córdoba (21)	0.164				0.471			0.031	Increasing*	0.447
Granada (21)	0.095	0.264	0.247	NS	0.272			NS		
Jaén (21)	0.117	0.369	0.100	NS	0.447			Increasing*	51	4
León (21)	0.014*				0.436			Increasing*	21	2
Madrid (21)	0.0007**				0.045			NS		
Málaga (21)	0.0002**				0.038			NS		
Ourense (21)	0.218				0.714	0.000	Increasing**	0.659	Increasing**	190
Santiago (21)	0.027*				−0.207	0.391	0.535	NS		
Girona (19)	0.933				0.575			0.010	Increasing*	0.481
Lleida (19)	0.240	0.419	0.074	NS	0.443			Increasing*	133	5
Manresa (19)	0.863	0.448	0.054	NS	0.423			Increasing*	266	4
Tarragona (19)	0.141	0.594	0.007	Increasing**	0.508			Increasing*	247	6
Vielha (11)	0.004**				0.528			0.536	0.709	NS
Toledo (10)	0.474				0.246	0.494	NS			0.055
Sta. Cruz Tenerife (10)	0.333	0.177	0.624	NS	−0.007	NS				
Izaña (9) [#]	0.003**				0.434	0.600	0.783	NS		
Palma de Mallorca (9)	0.696				0.151			0.697	NS	0.150
Roquetes-Tortosa (9)	0.031*				0.500			NS		
Burgos (8)	0.960				0.323			0.436	NS	0.286
Miranda (8)	0.079	−0.009	0.982	NS	0.238	0.643	0.833	NS		
Salamanca (8)	0.00009**				0.595			NS		
Segovia (8)	0.732				0.766			0.027	Increasing*	0.786
Soria (8)	0.483	−0.504	0.202	NS	−0.714			Decreasing*	−106	−4
Valladolid (8)	0.067	0.786	0.021	Increasing*	0.738			Increasing*	51	4
Almería (7)	0.027*				−0.036			0.714	0.893	NS
Arenas de San Pedro (7)	0.146				−0.552	0.199	NS			−0.357

Results of Shapiro–Wilk Normality, Pearson's correlation, Spearman's Rho tests and estimation of the magnitude. * Significance at 5%, ** significance at 1%; [#] the study period is from May to October; ¹ Shapiro–Wilk Normality Test. Reject H_0 : “there is normality” when p value $< 0.05^*$; $< 0.01^{**}$; ² Pearson's correlation only for stations with normality. When ρ value $< 0.05^*$; $< 0.01^{**}$, accept H_1 : “there is an increasing trend” when ρ value < 0.05 if $r > 0$; or H_1 : “there is a decreasing trend” when ρ value < 0.05 if $r < 0$; ³ Critical values (Zar 1972). When $|\rho| > C1$, accept H_1 : “there is an increasing trend” if $\rho > 0$; or H_1 : “there is a decreasing trend” when $|\rho| > C1$ if $\rho < 0$; ⁴ Theil–Sen estimator of the API trend (in number of pollen grains) calculated if PC or SP tests show a significant monotonic trend

absolute pollen peak varied from 6% (Madrid) and 7% (León) to 39% (Jaén) and 38% (Izaña) and accounted for 9–24% in the rest of the stations.

Figure 1b shows the pattern of the mean weekly pollen concentrations in the sampling stations during the study period. Clear differences in the

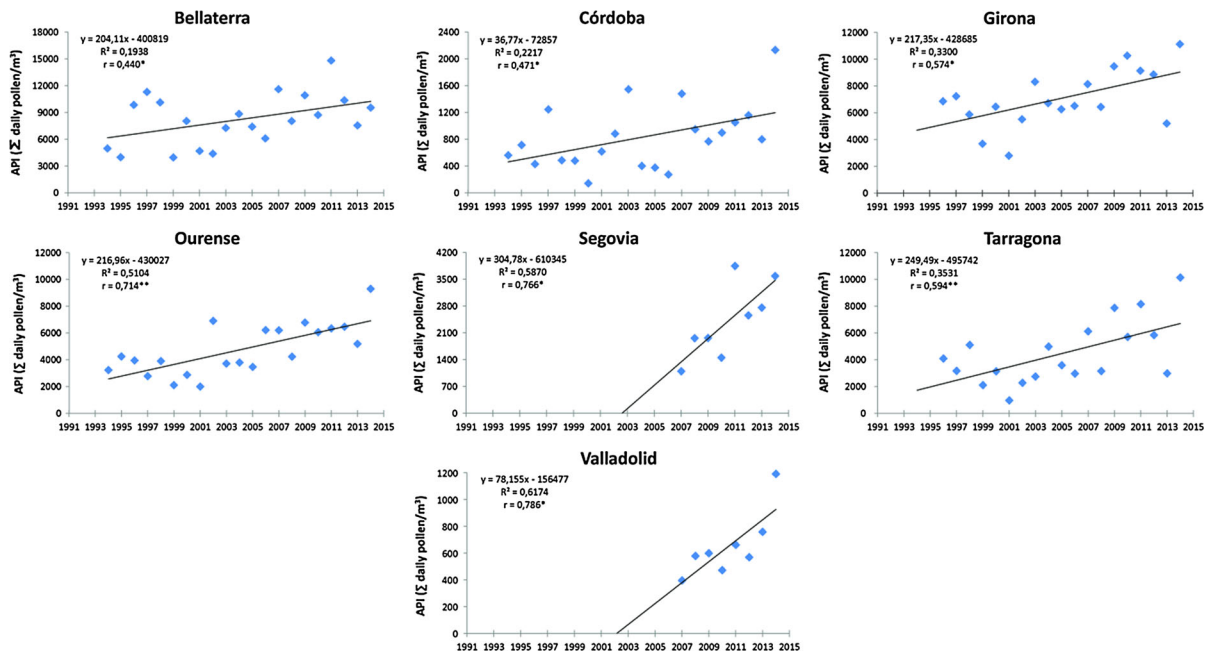


Fig. 3 Annual Pollen Index trends of *Pinus* along the studied period for aerobiological stations with normality distribution and significant Pearson correlation test

concentrations during the year between stations were detected, as indicated the diverse Y-axis scales ranging from 2500 to 20 pollen/m³. Figure 1b shows three pollination phases: the first from February to April, the second from April to June and the third with a single pollination phase.

3.2 Monotonical trends and correlation analysis

Table 3 summarizes the results of the statistical analysis carried out with *Pinus* API. SW test indicated that a total of 12 aerobiological stations showed non-normality (Almería, Badajoz, Barcelona, Cartagena, Izaña, León, Madrid, Málaga, Roquetes-Tortosa, Santiago, Salamanca and Vielha). PC and SR showed significant increasing trends in 7 stations: Bellaterra, Córdoba, Girona, Tarragona, Segovia, Valladolid (at 5%) and Ourense (at 1%), while some others showed significant increasing trends by using SR: Jaén, León, Lleida, Manresa (at 5%) and Cartagena (at 1%), and significant decreasing trends were found in Badajoz (at 1%) and Soria (at 5%). Theil–Sen estimators varied between an increase in the API ranging from 285 pollen grains (12%) in Segovia to 21 pollen grains (2%) in León, and a decrease of 106 pollen grains

(–4%) in Soria and 24 pollen grains (–4%) in Badajoz.

Scatterplots of the pollen time series with normality and significant trend, the corresponding regression equations with respect to the time and the coefficients of determination are displayed in Fig. 3. The highest linear regressions (1%) correspond to Ourense and Tarragona (increasing). Valladolid and Segovia (with the shortest sampling periods) showed the highest increasing trends (5%), followed by Girona, Córdoba and Bellaterra.

The MPI (Monthly Pollen Index) from January to August are shown in Table 4. January presented increasing MPIs in localities of the Castilla y León (León, Miranda de Ebro, Burgos, Valladolid, Segovia), in the South (Córdoba, Granada) and the eastern area (Cartagena, Tarragona, Palma de Mallorca). February kept the same trend in some localities of the Castilla y León (León, Valladolid, Segovia), although it showed an increase in Vielha and a decrease in littoral localities of Barcelona, Bellaterra, Tarragona and the inland area of Toledo, and Badajoz. March showed few significant correlations, resulting in decreases in Madrid and Santiago de Compostela and one increase in Cartagena, where April and May

Table 4 Spearman's Rho correlation between *Pinus* Monthly Pollen Indices (MPI) and years studied

Aerobiological station (years under study)	Spearman's Rho correlation ¹									
	ρ statistic								Critical values C1	
	January	February	March	April	May	June	July	August	5%	1%
Badajoz (21)	0.298	-0.410*	-0.643**	-0.532**	0.328	-0.024	-0.055	-0.341	0.370	0.508
Barcelona (21)	-0.046	-0.591**	-0.121	0.303	0.484*	0.303	0.142	0.050		
Bellaterra (21)	0.002	-0.533**	0.164	0.565**	0.459*	0.199	0.116	-0.193		
Cartagena (21)	0.545**	0.097	0.439*	0.415*	0.402*	0.337	0.394*	0.638**		
Córdoba (21)	0.624**	-0.151	0.064	0.055	0.688**	0.230	0.806**	0.480*		
Granada (21)	0.704**	-0.158	0.124	0.615**	0.402*	0.020	0.129	0.158		
Jaén (21)	0.749**	-0.002	0.224	0.704**	0.755**	0.167	0.076	-0.212		
León (21)	0.621**	0.537**	0.063	0.106	0.224	0.304	0.291	0.364		
Madrid (21)	0.312	0.245	-0.415*	0.038	-0.010	0.198	0.415*	0.326		
Málaga (21)	0.338	-0.084	-0.151	0.151	0.490*	0.228	0.463*	0.254		
Ourense (21)	-0.302	-0.159	-0.138	0.726**	0.481*	0.528**	0.746**	0.372*		
Santiago (21)	0.145	0.117	-0.450*	0.264	0.439*	0.639**	0.495*	0.175		
Girona (19)	-0.260	-0.123	0.327	0.265	0.258	0.455*	0.288	-0.036	0.391	0.535
Lleida (19)	-0.181	-0.318	-0.264	0.585**	0.418*	0.239	0.172	0.272		
Manresa (19)	0.111	-0.306	-0.234	0.460*	0.315	0.406*	0.260	-0.072		
Tarragona (19)	0.406*	-0.437*	0.222	0.393*	0.420*	0.388	0.297	-0.037		
Vielha (11)	-0.337	0.564*	0.382	0.500	0.373	0.255	0.219	0.173	0.536	0.709
Toledo (10)	0.285	-0.722*	0.358	0.237	0.431	-0.297	0.261	0.091	0.564	0.745
Sta. Cruz Tenerife (10)	0.407	-0.419	0.091	0.043	0.067	0.007	0.067	0.104	0.600	0.783
Izaña (9) [#]					-0.317	0.034	0.017	-0.517		
Palma de Mallorca (9)	0.667*	0.434	0.234	-0.350	0.467	-0.650*	-0.184	-0.467		
Roquetes-Tortosa (9)	0.234	0.117	0.317	0.350	0.367	0.434	0.234	-0.634*		
Burgos (8)	0.786*	-0.143	-0.334	-0.286	0.310	-0.048	-0.072	0.120	0.643	0.833
Miranda (8)	0.929**	0.191	-0.191	0.739*	-0.262	0.191	0.048	0.739*		
Salamanca (8)	0.334	0.286	-0.334	0.524	0.739*	0.310	0.905**	0.810*		
Segovia (8)	0.834**	0.786*	0.239	0.215	0.620	0.358	0.239	0.310		
Soria (8)	0.500	0.072	-0.620	-0.215	-0.477	-0.191	-0.262	0.072		
Valladolid (8)	0.643*	0.905**	0.024	0.334	0.691*	-0.191	0.667*	0.524		
Almería (7)	0.543	0.372	-0.429	-0.029	-0.772*	-0.658	0.143	-0.772	0.714	0.893
Arenas de San Pedro (7)	0.636	0.465	-0.536	-0.322	0.250	0.215	0.786*	0.393		

* Significance at 5%, ** significance at 1%. [#] the study period is from May to October. ¹ Critical values (Zar 1972). When $|p| > C1$, accept H_1 : "there is an increasing trend" if $p > 0$; and H_1 : "there is a decreasing trend" if $p < 0$

maintained the trend. April and May were the months with growing MPI values. In Manresa and Miranda de Ebro grew only in April, while in Barcelona (Catalonia), Córdoba, Málaga and Almería (Andalucía), and Salamanca and Valladolid (Castilla y León) only increased in May. Other Catalan localities (Bellaterra,

Lleida and Tarragona), Murcia (Cartagena) and Andalusian (Granada and Jaén) increased during both months. In the Atlantic North-western localities of Ourense and Santiago de Compostela, April–August and May–July, respectively, presented increasing MPI. June was the month than only showed significant

changes in Ourense and Santiago de Compostela (North-West) and in Girona and Manresa (North-East), where MPI increased, and in the Balearic islands (Palma de Mallorca) where it did the opposite. July and August tended to contain more *Pinus* pollen suspended in the air in Cartagena, Córdoba and Salamanca; only July in Madrid, Málaga, Valladolid and Arenas de San Pedro; and only August in Miranda de Ebro. In Roquetes-Tortosa, August MPI is decreasing. Finally, three localities (Santa Cruz de Tenerife and Izaña in the Canary Islands and Soria) did not show any trend in the MPI values.

The significant correlations between API and seasonal meteorological variables (Tmax, Tmin, Tmean and Prec) are shown in Table 5. It has been considered that the autumn and winter meteorological parameters had an important influence on the flowering (as well as API) of the next year. The winter and spring precipitations showed positive and significant correlations, especially in the stations with longest dataset.

4 Discussion

In recent years, several studies emphasized the vulnerability of forests to environmental change impacts, linking it to climate change (FAO 2012) or to biological and anthropological facts (Lenoir et al. 2010). Assuming that the use of a monitoring system capable of integrating climate and vegetation is necessary to understand this interaction, aerobiology is a good tool (Lewis et al. 2008). Because *Pinus* is one of the most dominant genera in the Northern Hemisphere forests, the study of long-term data series of airborne *Pinus* pollen in 30 Spanish localities offers the possibility to assess whether there are changes in the API trends and how they could affect in the ecological distribution of this taxon.

The *Pinus* API obtained showed differences between the studied areas. These variations were related with several factors such as coniferous forest cover, topography and species diversity. The stations located in Catalonia region showed the highest pine

Table 5 Spearman's Rho correlation between API and seasonal meteorological parameters

Aerobiological station (years under study; n)	Tmax Autumn (r^{-1})	Tmax Winter (r^{-1})	Tmax Summer	Tmean Autumn (r^{-1})	Tmean Winter (r^{-1})	Tmean Spring	Tmean Summer	Tmin Autumn (r^{-1})	Tmin Spring	Tmin Summer	Prec Winter (r^{-1})	Prec Spring	Prec Summer	Critical values C1
Badajoz (21)														5% = 0.370 1% = 0.508
Barcelona (21)									0.497*		0.544**	0.449*		
Bellaterra (21)											0.544**	0.449*		
Cartagena (21)										-0.604**				
Córdoba (21)		-0.481*				0.550**							0.465*	
Granada (21)					0.581**	0.638**	0.702**				0.517**	0.512**	0.766**	
Jaén (21)														
León (21)														
Madrid (21)														
Málaga (21)											0.663**			
Ourense (21)												-0.573**		
Santiago de Compostela (21)														5% = 0.391 1% = 0.535
Girona (19)								0.643**	0.481*	0.509*				
Lleida (19)	0.593**	0.593**												
Manresa (19)											0.434*			
Tarragona (19)											0.645**			5% = 0.536 1% = 0.709
Vielha (11)														
Toledo (10)						-0.759**								
Sta. Cruz Tenerife (10)														
Roquetes-Tortosa (9)											0.805**			5% = 0.600 1% = 0.783
Izaña (9) [#]	0.750*	0.750*						0.750*						
Palma de Mallorca (9)	0.750*	0.750*		0.780*				0.893**		-0.690*				
Burgos (8)	0.786*													
Miranda de Ebro (8)														5% = 0.643 1% = 0.833
Salamanca (8)	0.811*					-0.731*								
Segovia (8)	0.775*													
Soria (8)														
Valladolid (8)														5% = 0.714 1% = 0.893
Almería (7)			-0.806*											
Arenas de San Pedro (7)														

* Significance at 5%, ** significance at 1%. [#] the study period is from May to October. ¹Critical values (Zar 1972). When $|p| > C1$, accept H_1 : "there is an increasing trend" if $p > 0$; or H_1 : "there is a decreasing trend" if $p < 0$

pollen concentrations range in Roquetes-Tortosa (10,598 pollen grains per year) and the lowest in Lleida (2889 pollen grains). Canary Islands represented the opposite case, showing the lowest *Pinus* API registered (122 in Santa Cruz de Tenerife and 251 in Izaña). The forest cover plays an important role in these concentrations. E.g. Badajoz presents landscapes with low *Pinus* coverage, only a 4% of the total forest of this area corresponds to conifers (DGCN, IFN3 2007), and showed a low API (578 pollen grains, around 1% of the total pollen collected). In Galicia region, Santiago de Compostela area presents a 24% of coniferous taxa of the total forest cover and registered 780 pollen grains, while Ourense area, with a total coniferous cover of 47% of the province (DGCN, IFN3 2007), registered 4760 pollen grains. In Andalusia region, Córdoba registered the lowest concentrations (831 pollen grains) while the rest stations were over 1040 pollen grains. In this case, in all the Andalusian provinces studied, the coniferous cover is over 40% of the total forest except Córdoba, which has 11%. In this study, the three stations placed in mountains zones registered high pollen values: Roquetes-Tortosa with the maximum API of study, and Vielha (Catalonia), and Arenas de San Pedro (Castilla y León).

The absolute daily maximum concentration showed the possible origin of the pollen in the area studied, e.g., the sampling stations with the highest absolute values were Jaén and Izaña. In both cases near to 40% of the total *Pinus* pollen were collected in one day. According to Green et al. (2003) that showed the great capability of *Pinus* pollen to be transported by air masses, this result suggests that the pollen recorded in both stations could proceed of remote areas, causing punctual events of long-range transport. Furthermore, this phenomenon may also be observed during the pollination period from May to July. The aerobiological stations such as Almería, Bellaterra, Barcelona, Girona, Granada, Jaén, León, Madrid, Manresa, Miranda de Ebro, Segovia, Salamanca or Castilla-La Mancha showed an increase in pollen concentration in the late spring and first of summer (Fig. 1b). This increase could be related with airborne pollen release by pine species located in remote mountains areas and afterward transported for long distances. However, the increase of pollen in May was also in Cartagena, Córdoba, Palma de Mallorca stations. In these cases, where *P. nigra*, *P. sylvestris* or *P. uncinata* are not

present, the responsible could be the extensive blooms of the *Pinus* species used as ornamental in the urban areas.

Considering the yearly dynamics of the mean weekly *Pinus* pollen concentrations, different patterns have been detected. As already observed in other European localities (Jato et al. 2000; Frenguelli et al. 2002), *Pinus* pollen showed two main pollination phases (February–May and May–June) depending on the species distribution and flowering. In general, according to the *Pinus* species distribution and the yearly dynamics of API in each studied (Fig. 1b), we can speculate the flowering period of the species. *P. halepensis*, *P. pinea* and *P. pinaster* (species at the littoral and lower territories) that show lower sensitivity to the winter temperatures (Frenguelli et al. 2002) and to the summer drought, started in the first months of the year, accordance with the Flora Iberica (Castroviejo et al. 1993). In the regions of Extremadura, Murcia, the Balearic Islands, Andalusia, the flowering started in February while in Castilla y León, Catalonia, Madrid, Castilla-La Mancha began in March. The flowering of *P. pinaster* starts in February (Galicia region) and finished in June (Madrid, Castilla-La Mancha, Córdoba, Jaén, Málaga). However, the specie of the mountain (*P. sylvestris*) and subalpine (*P. uncinata*) areas, the cooler temperatures and the water availability during summer (Sabaté et al. 2002; Thuiller et al. 2003) provoke a late flowering. Thirteen stations presented a single pollination phase. Santiago de Compostela, Ourense, Málaga and Jaén registered airborne *Pinus* pollen from February to April due to the earliest flowering in *P. halepensis*, *P. pinaster*, *P. pinea*, *P. radiata* which are considered littoral and low land areas species. The aerobiological stations of Castilla y León (León, Valladolid, Salamanca, Segovia, Soria, Arenas de San Pedro) showed also a single pollination period from April to June due to the species of low land areas. In this region, the flowering is later due to the meteorological conditions (with colder winter than Galicia or Andalusia regions), as well as to the mountain specie of *P. sylvestris* that started in May and overlap with the others species. Another station with one pollination phase was Vielha (Pyrenean valley) which was represented by *P. sylvestris* and *P. uncinata*, and registered airborne *Pinus* pollen from May to July. Finally, Santa Cruz de Tenerife and Izaña, located in the volcanic island of Tenerife, recorded *Pinus* pollen from the endemic *P.*

canariensis and from the silvicultured *P. radiata*. As previously described (Navascués et al. 2007), the dramatic difference in altitude in Tenerife (0–3718 m) induces asynchronous flowering times among elevations. It can explain the continuous presence during the whole years of pollen in Santa Cruz de Tenerife, at 37 m.a.s.l. In Izaña, at 2367 m.a.s.l. the only pollination peak recorded is due to the late beginning of flowering in May as well as to the closest *Pinus* trees; the pollen quantity registered was four times more of the concentration in Santa Cruz de Tenerife. The trends analysis obtained for the flowering intensity of *Pinus* (Table 4) showed that the starting of the flowering was advancing while the end occurred later, and so the pollination period was lengthening.

Regarding the statistical analysis, 14 stations showed significant monotonic trend: 12 increasing with significant trends that can suppose an annual increase between 12% (Segovia) to 2% (León). Only two stations showed significant decreasing trends (Soria and Badajoz with an annual decrease of −4%). There is a general consensus among scientists that the climate changes observed in the last century are affecting the forest cover. In fact, the comparison of the data of Allué (1990) with the meteorological data of the studied period (1994–2014) suggests relevant climatological differences. In general, the study suggests a significant increase of the pollen trends for the whole registered stations, as well as the mean temperature, in contrast to the amount of rainfall. However, it is not an evident cause of the increasing trend in the pollination of *Pinus* because in two stations (Badajoz and Soria) with a decreasing trend or other stations without significant trends, the climatic changes have been similar.

Forests are particularly sensitive to environmental changes because of their low adaptations capacity to several factors (Dale et al. 2001; Lindner et al. 2010). Some studies on European forest ecosystems found interesting differences in the response to these changes. Some authors postulate an increase in the coniferous forest growth in Northern Europe (Briceño-Elizondo et al. 2006; García-Gonzalo et al. 2007), and, on the other hand, a negative impacts were shown in the Southern and Eastern regions, with a Mediterranean or continental influence (Benito Garzón et al. 2008; Lindner et al. 2010; Attorre et al. 2011; Sánchez-Salguero et al. 2012). Furthermore in Spain two different trends have been found between North

and South areas. The first was detected in Catalonia where the growth of *P. sylvestris* has been favored in the last decades (Martínez-Vilalta et al. 2008), as well as *P. uncinata* has adapted its morphology and physiology in order to colonize higher territories (Ameztegui and Coll 2011). The second was determined in Andalusia by the species of *P. nigra* and *P. sylvestris* which have suffered a decline process (Linares and Tiscar 2010; Sánchez-Salguero et al. 2012). Our data supported observations postulated in Catalonia because the *Pinus* API showed a significant increasing trend in five stations (Bellatera, Girona, Lleida, Manresa, Tarragona), unlike what happened in Southern Spain. Although Badajoz and Almeria showed decreasing trends (only significant in the first case), Córdoba, Jaén, Cartagena, Granada and Málaga showed increasing trends (significant in the three first cases).

The significant decrease of the trends in some stations could be related to other factors such as forest fires, species interactions, pollen dispersal capacity, abandonment of silviculture, or habitat modification. Future studies will be conducted in order to clarify these relationships.

Despite the absence of a general behavior, winter and spring precipitations were the variables with the highest influence in the stations with longest datasets, when considering API and seasonal meteorological data. Daily data should be confronted in order to better understand the relationship between them. According to Jato et al. (2000), the morphology of the pollen grains of *Pinus* could facilitate their persistence within the atmosphere once they have been freed, so they are not affected by the temperature and precipitation during the blooming period.

5 Conclusions

Our findings show that airborne pollen data series are good bio-indicators of changes happening in *Pinus* trees landscapes, and therefore could explain the vulnerability of the different species present in Spain.

While the littoral and low land pine species (*P. halepensis*, *P. pinaster*, *P. pinea*, *P. radiata*) have a clear influence in the outline of the airborne pollen patterns during the first months of the year, *Pinus sylvestris* and *P. uncinata* (mountain species) had an influence in the second pollination phase (May to June).

The flowering period of *Pinus* in Spain is changing, with increases in their concentrations at the beginning and end of period and overlapping the flowering of the littoral and low pine species with those of the mountain areas and subalpine regions.

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References

- Alba-Sánchez, F., Sabariego-Ruiz, S., Díaz de la Guardia, C., Nieto-Lugilde, D., & De Linares, C. (2010). Aerobiological behaviour of six anemophilous taxa in semi-arid environments of southern Europe (Almería, SE Spain). *Journal of Arid Environment*, 74, 1381–1391.
- Allué, J. L. (1990). *Atlas fitoclimático de España [Phytoclimatic Atlas of Spain]*. Madrid: INIA.
- Ameztegui, A., & Coll, L. (2011). Tree dynamics and co-existence in the montane-sub-alpine ecotone: The role of different light-induced strategies. *Journal Vegetation Science*, 22, 1049–1061.
- Attorre, F., Alfo, M., De Sanctis, M., Francesconi, F., Valenti, R., Vitale, M., et al. (2011). Evaluating the effects of climate change on tree species abundance and distribution in the Italian peninsula. *Applied Vegetation Science*, 14(2), 242–255.
- Belmonte, J., Alarcón, M., Avila, A., Scialabba, E., & Pino, D. (2008). Long-range transport of beech (*Fagus sylvatica* L.) pollen to Catalonia (north-eastern Spain). *International Journal of Biometeorology*, 52(7), 675–687.
- Benito Garzón, M., Sánchez de Dios, R., & Sainz Ollero, H. (2008). Effects of climate change on the distribution of Iberian tree species. *Applied Vegetation Science*, 11(2), 169–178.
- Briceño-Elizondo, E., García-Gonzalo, J., Peltola, H., Matala, J., & Kellomaki, S. (2006). Sensitivity of growth of Scots pine, Norway spruce and silver birch to climate change and forest management in boreal conditions. *Forest Ecology Management*, 232, 152–167.
- Cariñanos, P., De la Guardia, C., Algarra, J. A., De Linares, C., & Irurita, J. M. (2013). The pollen counts as bioindicator of meteorological trends and tool for assessing the status of endangered species: The case of *Artemisia* in Sierra Nevada (Spain). *Climatic Change*, 119, 1–15.
- Cariñanos, P., Galán, C., Alcázar, P., & Domínguez, E. (2004). Airborne pollen records response to climatic conditions in arid areas of the Iberian Peninsula. *Environmental and Experimental Botany*, 52, 11–22.
- Cariñanos, P., Galán, C., Alcázar, P., & Domínguez, E. (2010). Airborne pollen records and status of the anemophilous flora in arid areas of the Iberian Peninsula. *Journal of Arid Environments*, 74, 1102–1105.
- Castroviejo, S., Aedo, C., Cirujano, S., Laínz, M., Montserrat, P., Morales, R., et al. (Eds.). (1993). *Flora ibérica I*. Madrid: Real Jardín Botánico, CSIC.
- Clot, B. (2003). Trends in airborne pollen: An overview of 21 years of data in Neuchâtel (Switzerland). *Aerobiologia*, 19, 227–234.
- Dale, V. H., Joyce, L. A., McNulty, S., & Neilson, R. P. (2000). The interplay between climate change, forests, and disturbances. *Science of the Total Environment*, 262, 201–204.
- Dale, V. H., Joyce, L. A., McNulty, S., Neilson, R. P., Ayres, M. P., Flannigan, M. D., et al. (2001). Climate change and forest disturbances. *BioScience*, 51(9), 723–734.
- DGCN, IFN3. (2007). *Tercer Inventario Forestal Nacional, IFN3 (1997–2007) [Third National Forest Inventory, IFN3 (1997–2007)]*. Madrid: Ministerio de Medio Ambiente. <http://www.magrama.gob.es/es/biodiversidad/servicios/banco-datos-naturaleza/informacion-disponible/ifn3.aspx>. Accessed November 9, 2016.
- Docampo, S., Recio, M., Trigo, M. M., Melgar, M., & Cabezudo, B. (2007). Risk of pollen allergy in Nerja (southern Spain): A pollen calendar. *Aerobiologia*, 23, 189–199.
- FAO. (2011). *State of the world's forests*. Rome: Food and Agriculture Organization of the United Nations.
- FAO. (2012). *Forest management and climate change: A literature review*. Rome: Food and Agriculture Organization of the United Nations.
- Fernández-González, M., Rodríguez-Rajo, F. J., Jato, V., Escuredo, O., & Aira, J. M. (2011). Estimation of yield “Loureira” variety with an aerobiological and phenological model. *Grana*, 50, 63–72.
- Fernández-Llamazares, A., Belmonte, J., Delgado, R., & De Linares, C. (2014). A statistical approach to bioclimatic trend detection in the airborne pollen records of Catalonia (NE Spain). *International Journal of Biometeorology*, 58, 371–382. doi:10.1007/s00484-013-0632-4.
- Frei, T., & Gassner, E. (2008). Climate change and its impact on birch pollen quantities and the start of the pollen season an example from Switzerland for the period 1969–2006. *International Journal of Biometeorology*, 52, 667–674.
- Frenguelli, G., Tedeschini, E., Veronesi, F., & Bricchi, E. (2002). Airborne pine (*Pinus* spp.) pollen in the atmosphere of Perugia (Central Italy): Behaviour of pollination in the two last decades. *Aerobiologia*, 18, 223–228.
- Galán, C., Alcázar, P., Oteros, J., García-Mozo, H., Aira, M. J., Belmonte, J., et al. (2016). Airborne pollen trends in the Iberian Peninsula. *Science of the Total Environment*, 550, 53–59.
- Galán, C., Cariñanos, P., Alcázar, P., & Domínguez, E. (2007). *Manual de Calidad y Gestión de la Red Española de Aerobiología [Quality manual and management Spanish aerobiology network]*. Spain: Servicio de Publicaciones de la Universidad de Córdoba.

- Galán, C., Smith, M., Thibaudon, M., Frenguelli, G., Oteros, J., Gehrig, R., et al. (2014). Pollen monitoring: Minimum requirements and reproducibility of analysis. *Aerobiologia*, 30(4), 385–395.
- García-Gonzalo, J., Peltola, H., Briceño-Elizondo, E., & Kellomäki, S. (2007). Changed thinning regimes may increase carbon stock under climate change: A case study from a Finnish boreal forest. *Climatic Change*, 81, 431–454.
- García-Mozo, H., Mestre, A., & Galán, C. (2010). Phenological trends in southern Spain: A response to climate change. *Agricultural Forest Meteorology*, 150, 575–580.
- Gobierno de Canarias. (2007) Programa de desarrollo rural de Canarias. FEADER (2007) [Rural Development Programme de Canarias. FEADER 2007]. Canarias: Gobierno de Canarias. <http://www.pdrcanarias.org>. Accessed November 9, 2016.
- Green, B. J., Panula, E. Y., Dettmann, M., Rutherford, S., & Simpson, R. (2003). Airborne *Pinus* pollen in the atmosphere of Brisbane, Australia and relationships with meteorological parameters. *Aerobiologia*, 19, 47–55.
- Hirst, J. M. (1952). An automatic volumetric spore-trap. *Annals of Applied Biology*, 38, 257–265.
- Jato, M. V., Rodríguez, F. J., & Seijo, M. C. (2000). *Pinus* pollen in the atmosphere of Vigo and its relationship to meteorological factors. *International Journal of Biometeorology*, 43, 147–153.
- Lenoir, J., Gégout, J. C., Guisan, A., Vittori, P., Wohlgemuth, T., Zimmermann, N. E., et al. (2010). Going against the flow: Potential mechanisms for unexpected downslope range shifts in a warming climate. *Ecography*, 33(2), 295–303.
- Lewis, H., Ziska, L. H., Epstein, P. R., & Rogers, C. A. (2008). Climate change, aerobiology, and public health in the Northeast United States. *Mitigation and Adaptation Strategies for Global Change*, 13, 607–613. doi:10.1007/s11027-007-9134-1.
- Linares, J. C., & Tiscar, P. A. (2010). Climate change impacts and vulnerability of the southern populations of *Pinus nigra* subsp. *salzmannii*. *Tree Physiology*, 30(7), 795–806.
- Lindner, M., Maroschek, M., Netherer, S., Kremer, A., Barbati, A., García-Gonzalo, J., et al. (2010). Climate change impacts, adaptive capacity, and vulnerability of European forest ecosystems. *Forest Ecology Management*, 259, 698–709.
- Martínez-Vilalta, J., López, B. C., Adell, N., Badiella, L., & Ninyerola, M. (2008). Twentieth century increase of Scots pine radial growth in NE Spain shows strong climate interactions. *Global Change Biology*, 14, 2868–2881.
- Navascués, M., Vendramin, G. G., & Emerson, B. C. (2007). The effect of altitude on the pattern of gene flow in the endemic canary island pine, *Pinus canariensis*. *Silvae Genetica*, 57, 357–363.
- Pearson, K. (1896). Mathematical contributions to the theory of evolution.—on a form of spurious correlation which may arise when indices are used in the measurement of organs. *Proceedings of the Royal Society of London*, 60(359–367), 489–498.
- Pérez Badia, R., Rapp, A., Morales, C., Sardinero, S., Galán, C., & García-Mozo, H. (2010). Pollen spectrum and risk of pollen allergy in central Spain. *Annals of Agricultural and Environmental Medicine*, 17, 139–151.
- R Development Core Team. (2007). *R: A language and environment for statistical computing*. Vienna: R Foundation for Statistical Computing.
- Rodríguez-Rajo, F. J., Jato, V., & Aira, M. J. (2003). Pollen content in the atmosphere of Lugo (NW Spain) with reference to meteorological factors (1999–2001). *Aerobiologia*, 19, 213–225.
- Rosenzweig, C., Karoly, D., Vicarelli, M., Neofotis, P., Wu, Q., Casassa, G., et al. (2008). Attributing physical and biological impacts to anthropogenic climate change. *Nature*, 453, 353–358.
- Ruiz García, L., Díaz de la Guardia, C., & Mota, J. F. (1998). Analysis of airborne pollen in the town of Almería (South-East Spain), 1995–1996. *Aerobiologia*, 14, 281–284.
- Sabaté, S., Gracia, C. A., & Sánchez, A. (2002). Likely effects of climate change on growth of *Quercus ilex*, *Pinus halepensis*, *Pinus pinaster*, *Pinus sylvestris* and *Fagus sylvatica* forests in the Mediterranean region. *Forest Ecology Management*, 162, 23–37.
- Sánchez-Salguero, R., Navarro-Cerrillo, R. M., Swetnam, T. W., & Zavala, M. A. (2012). Is drought the main decline factor at the rear edge of Europe? The case of southern Iberian pine plantations. *Forest Ecology Management*, 271, 158–169.
- Sen, P. K. (1968). Asymptotic normality of sample quantiles for m -dependent processes. *Annals of Mathematical Statistics*, 39, 1724–1730.
- Shapiro, S. S., & Wilk, M. B. (1965). An analysis of variance test for normality (complete samples). *Biometrika*, 52(3/4), 591–611.
- Spearman, C. (1904). “General Intelligence” objectively determined and measured. *American Journal of Psychology*, 15(2), 202–286.
- Theil, H. (1950). A rank-invariant method of linear and polynomial regression analysis. I, II, III. *Proceedings of the Koninklijke Nederlandse Academie van Wetenschappen*, 53, 386–392.
- Thuiller, W., Vayreda, J., Pino, J., Sabate, S., Lavorel, S., & Gracia, C. (2003). Large-scale environmental correlates of forest tree distributions in Catalonia (NE Spain). *Global Ecology Biogeography*, 12, 313–325.
- Tormo-Molina, R., Gonzalo-Garijo, M. A., Silva-Palacios, I., & Muñoz-Rodríguez, A. F. (2010). General trends in airborne pollen production and pollination periods at a Mediterranean site (Badajoz, Southwest Spain). *Journal of Investigational Allergology and Clinical Immunology*, 20(7), 567–574.
- Tormo-Molina, R., Silva-Palacios, I., Gonzalo-Garijo, A., Moreno-Corcheró, A., Pérez-Calderón, R., & Fernández-Rodríguez, S. (2011). Phenological records as a complement to aerobiological data. *International Journal of Biometeorology*, 55, 51–65.
- Womack, A. M., Bohannon, B. J. M., & Green, J. L. (2010). Biodiversity and biogeography of the atmosphere. *Philosophical Transactions of the Royal Society of London*, 365, 3645–3653.
- Zar, J. H. (1972). Significance testing of the spearman rank correlation coefficient. *Journal of the American Statistical Association*, 67, 339.
- Ziello, C., Sparks, T. H., Estrella, N., Belmonte, J., Bergmann, K. H., Bucher, E., et al. (2012). Changes to airborne pollen counts across Europe. *PLoS ONE*, 7, 1–8.