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1 HYSPLIT as an environmental impact assessment tool to study the data

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1 Abstract

2 The olive tree (Olea europaea) is a native evergreen tree in the Mediterranean region, being one of the most important causes of seasonal respiratory allergies in Mediterranean countries. 3 This work aims to relate flowering phenology, source tree distribution, meteorology, and 4 5 airborne pollen records for this species and to analyse the possible arrival of air masses from distant areas during days when differences between the phenological and pollen peaks. 6 Aerobiological sampling was carried out in Badajoz (SW Spain) for 4 years (2016-2019) 7 using a Hirst volumetric sampler. Trees were geolocalized in the city and surrounding areas. 8 9 The pollination phenology of 15 specimens was studied for four years (2016-2019) during 10 the months from April to June. The daily data for the whole period and the hourly data for the four years, including pollen records and meteorology, were analysed. The comparison 11 between the pollen records and sources distribution was assessed. The main pollen season 12 (thereafter, MPS) lasted an average of 34.5 (29-40) days. Phenological observations indicate 13 that pollination occurred for 26.5 days and was mostly within the period of recorded airborne 14 pollen; however, were days with airborne pollen recorded outside the pollination period. In 15 2017 the peak day was recorded when the flowering period has reached only the 10%, in 16 other seasons this value reached the maximum peak of pollen concentration was found only 17 18 a few days after the maximum of flowering. The hourly analysis showed that the maximum pollen concentrations were reached just after noon. The analysis of pollen sources and pollen 19 records showed a close relationship with the predominant winds and tree distributions. The 20 observed discrepancies between phenological and aerobiological data (in 2017 and 2018) 21 were explained by the movement of air masses and long-distance transport. 22

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24 Keywords: Aerobiology, Botany, Phenology, HYSPLIT, Urban maps, Olea pollen

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26 **1.** Introduction

The monitoring of airborne olive tree pollen is very important from the points of view of agriculture, ecology (Rallo & Cuevas, 2001; Pérez-Badia, 2015a) and medicine (D'Amato *et al.* 2007; Salamanca *et al.*, 2010; Vara *et al.* 2016) due to the close relationship between fruit production, high allergenic potential, high pollen production by the tree and extensive cultivation (SEAIC, 2005; Rojo *et al.*, 2016). Although aerobiological studies have traditionally been applied to allergy research, their usefulness for crop prognosis is currently
being demonstrated, and they are highly desirable from an economic standpoint, for
harvesting and for planning olive oil marketing and global commercial distribution (Llerena
& Garrido, 2010; Galán *et al.*, 2004).

The olive tree is an entomophilous species that has evolved towards anemophily with 36 many flowers, and an adult olive tree can produce large quantities of pollen grains (Rojo et 37 al., 2016; Ferrara et al., 2007). The genus Olea includes approximately 35 species 38 worldwide; 98% of olive groves are concentrated in the Mediterranean area and 24% are in 39 40 Spain, mostly present in Andalucía (62%), Castilla-La Mancha (15%) and Extremadura 41 (11%), and consist of Olea europaea subsp. europaea var. europaea (Vargas & Talavera, 2012) as the most cultivated taxon and an important oil-producing crop (Sefc et al., 2001; 42 Rojo 2014). In Extremadura, five varieties dominate those planted (e.g., Manzanilla de 43 Sevilla, Manzanilla Cacereña, Cornicabra, Verdial de Badajoz, and Morisca), being the first 44 that was present in the study area (Llerena & Garrido, 2010). 45

Moreover, olive tree pollen is considered to be one of the main pollen types responsible for allergic diseases that occur in the Mediterranean region (D'Amato *et al.*, 2007). An elevated proportion of Mediterranean patients show poli-sensitizations that reach a rate of 80% within the group of *Olea* allergy sufferers (Moreno-Grau *et al.* 2016). In some regions of southern Spain, olive tree pollen is the main cause of allergic sensitization and is the cause of sensitization in more than 40-45% of sensitized individuals (Moreno-Grau *et al.* 2016; Salamanca *et al.*, 2010).

Furthermore, despite this fact, in recent years, there has been much planting of this
species in urban green spaces that produces large amounts of pollen (Staffolani *et al.*, 2011;
Galán *et al.*, 2016; Charalampopoulus *et al.*, 2018). Allergic problems in urban environments
should be considered in the design of the public environments (Velasco-Jiménez *et al.*, 2014),
being olive trees frequently used as ornamentals in urban environments that have allergenic
properties (Cariñanos & Casares-Porcel, 2011; Cariñanos *et al.*, 2014).
Allergenic properties are relevant to visitor behaviour and the characteristics of green spaces

in Spain (Adinolfi *et al.*, 2014; Mohammad & Pooryousef, 2011; Fernández-Rodríguez *et al.*

61 2018; Maya-Manzano *et al.*, 2017a).

Phenological studies are an important and complementary part of aerobiological 63 monitoring (Monroy-Colín et al. 2018), since they allow connecting emission sources with the airborne pollen records and enable better interpretation of the aerobiological results 64 (Fernández-Rodríguez et al. 2014a; Tormo et al., 2011); in addition to allowing the 65 population with allergenic problems to be alerted to the increase in pollen in the air (Bruns 66 et al. 2013; Carter et al. 2017; Monroy-Colín et al. 2018). Furthermore, the information about 67

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68 the flowering periods helps to differentiate species within the same pollen type by their pollen curves at a specific time (Monroy-Colín et al. 2018; Zerboni, 1998). 69

The olive tree blooms during the spring and flower production is conditioned by 70 71 important physiological regulations that depend directly on the environmental conditions 72 from the previous year (Rojo, 2014; Galán, et al., 2001a). In addition, there are genetic 73 conditions that are specific to each variety (Rosati et al., 2012; Rojo, 2014; Rojo & Pérez-74 Badia, 2015a) and the timing and intensity of the pollen curves are strongly influenced by meteorological parameters (Hernández-Ceballos et al., 2012). Olive tree flowering shows an 75 76 annual cycle, including bud formation during the previous summer, dormancy during the cold period, budburst in late winter, and flower structure development from budburst to 77 flowering in the spring (Aguilera et al. 2015a; Zhu, et al.; 2012). Temperature is one of the 78 79 main factors affecting the flowering of olive trees, and a low-temperature period prior to bud development is essential to interrupt dormancy (Aguilera et al. 2015a; Galán, et al., 2001a). 80 81 Light affects the induction of floral buds in olive trees, but its impact on the floral phenology of olive trees is much smaller than that of temperature, and these depend on olive tree 82 cultivars and geographical locations (Zhu, et al.; 2012; Galán, et al., 2001a). Several studies 83 have reported that the weather-related variable that is most influential for olive tree flowering 84 is the temperatures during the months prior to anthesis (Galán, et al. 2001a; Galán, et al., 85 86 2001b; Rojo & Pérez-Badia, 2015a). The floral phenological behaviours of olive trees are very consistent in response to similar meteorological conditions and are independent of 87 latitude variations (Orlandi, 2005). Olea pollen in the air is influenced by several factors, 88 such as time of day, season, weather conditions, geographical locations of sources 89 (Fernández-Rodríguez et al. 2016) and the vegetation type that dominates one area 90 (Charalampopoulus et al., 2018). 91

It is for this reason that detailed mapping of the vegetation must be available, from which 92 93 the pollen sources can be identified (Skjøth et al., 2013; Maya-Manzano et al., 2017b). Airborne pollen counts in areas with different levels of urbanization reveal differences in the 94 number of pollen types recorded (Fernández-Rodríguez et al. 2014b; Skjøth et al., 2013). 95 96 Therefore, to know the distributions of ornamental plants with potentially allergenic pollen and its phenology allows to create urban risk maps (Pecero-Casimiro et al. 2019) and to take 97 98 preventive measures that allows better preparation by hospital emergency services, which are often saturated during periods of high pollen levels (Galán, et al., 2001a; Charalampopoulus 99 et al., 2018). Also to reduce the pollen exposure and to initiate treatment at the appropriate 100 101 times (Monroy-Colín et al. 2018; Tedeschini et al., 2006). The pollen distributions within cities depend on various factors such as microclimates, the spatial locations of trees, the 102 predominant wind directions as related to source locations and the building heights (Maya-103 Manzano et al., 2017b; Charalampopoulus et al., 2018; Fernández-Rodríguez et al., 2018). 104 Geostatistical techniques and geographic information systems have been used recently for 105 106 modelling olive tree flowering. These tools provide reliable interpretations of aerobiological data and the factors involved in airborne pollen dispersal (Rojo & Pérez-Badia, 2015). 107 Recently, there has been an increase in the mapping of these trees in urban environments 108 109 (Maya-Manzano et al., 2017a) and in regional areas to model pollen intensities (Rojo et al., 2016; Aguilera et al., 2015b). 110

Based on previous aerobiological analyses in the study area (Fernández-Rodríguez et al., 111 2014a) and in nearby locations (Hernández-Ceballos et al., 2011a; Hernández-Ceballos et 112 al., 2014b; Hernandez-Ceballos et al., 2012), long-distance transport episodes should be 113 considered in the study of the temporal cycles of pollen concentrations and their attribution 114 to local sources (Skjøth et al., 2012). Furthermore, regional scale transport of olive tree pollen 115 116 can result in increased nightly concentrations of this important aeroallergen (Fernández-Rodríguez et al., 2014a). A number of studies have examined the long-distance transport 117 episodes (LDT) of airborne pollen grains and have attempted to identify their sources, e.g., 118 the pollen types from trees such as Betula spp. (Skjøth et al., 2014; Skjøth et al., 2009; Skjøth 119 et al., 2007); Quercus spp. (Hernández-Ceballos et al., 2011b; Hernández-Ceballos et al., 120 2014a; Maya-Manzano et al., 2016) and Olea spp. (Fernández-Rodríguez et al., 2014a; 121 122 Hernández-Ceballos et al., 2011a; Hernández-Ceballos et al., 2012; Hernández-Ceballos et

al., 2014b). In this sense, this mapping, combined with dispersion models as Hybrid Single 123 Particle Lagrangian Integrated Trajectory (HYSPLIT), (Draxler & Hess, 1998) can be helpful 124 for achieving a full understanding of some of the discrepancies between pollen content and 125 phenology. In this work, the backward dispersion calculation for particles was used, and it 126 127 provided information for the characteristics of air-mass movements over a region, in terms of their origin, horizontal pathways and altitudes. This information also provides detailed 128 129 information on the paths followed by air masses until its arrival over the study area and is therefore a useful tool for a range of scientific applications that are related to air-quality 130 131 analysis (Borge et al. 2007; Hernández-Ceballos et al. 2011b).

The aim of this study is to evaluate the relationships of airborne *Olea europaea* pollen recorded by the pollen trap and the arrangements of olive trees within the city of Badajoz (Spain), and to relate the influence of vegetation through the study of phenology to the presence of pollen in the air. Additionally, some LDT were analysed to determine the possible arrival of air masses from distant places during these divergences between the phenological and pollen peaks.

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2. <u>Material and methods</u> 2.1. <u>Study area</u>

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This study was conducted in Badajoz, a city in the SW of Spain with 150 543 inhabitants
(NSI, 2018). The city is 184 m above sea level and is crossed by the Guadiana River with the
Gévora River as a tributary.

Daily meteorological data were provided by the National Meteorology Agency (AEMET) 144 from a meteorological station located at 38° 53' 00" N, 6° 48' 50" W, which was 3.7 km from 145 the aerobiological sampler. This station provided data of Mean, maximum and Minimum 146 147 temperature (°C), rain $(1/m^2)$, wind direction (km/h) and wind direction (degrees). Moreover, for hourly data, a portable weather station (WS-GP1 Delta-T) was located 2 metres from the 148 pollen trap (Fig. 1A) to achieve more similar results to the pollen sampling point. The 149 portable meteorological station data were recorded every 10 minutes, and included 150 temperature (°C), rain (mm), relative humidity (%), wind speed (m s⁻¹) and wind direction 151 (grades) and provided data for the four years of the study. To process the wind direction data, 152 10-minute values were transformed into 30° sections (12 values) and the modes of the hourly 153

data were calculated; for the remaining meteorological parameters, average values were usedexcept for rain, for which the sums were calculated.

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2.2.*Olea* trees mapping

Olea europaea trees in the city were examined during the period of this study (2016 to 157 158 2019) and were counted and geo-referenced on a map (Fig. 1B) and included ornamental or urban green areas and the city outskirts. Also, the mapping for those specimens whose 159 160 phenology was studied have been included (Fig. 1B). Major olive tree pollen sources were identified using the Corine Land Cover (CLC) 2006 v. 17 datasets for the studied area (EC, 161 162 2013). The inventory maps were gridded using the CLC 2012 projection by aggregating the 163 grid cells to 20 km x 20 km using procedures similar to Sadyś et al., (2014) and Skjøth et al., (2012). This procedure enables easy comparisons of habitat densities at regional and national 164 scales. Olive trees appear as agricultural areas with permanent crops and are denoted as olive 165 tree groves (code 223). Furthermore, layers of the elevation data of the Iberian Peninsula 166 167 were used (Fig. 2).

168 2.3. Pollen sampling

Olea pollen grains were collected using a Burkard seven-day pollen trap (Hirst, 1952). 169 The trap was located on a 6 m high terrace and was placed in the Agricultural Engineering 170 171 School, in Badajoz at SW Iberian Peninsula (38°53' N, 6°58' W) (Fig. 1). Standardized data management procedures for capturing and counting airborne pollen were used, according to 172 173 the Spanish Aerobiology Network (REA) (Galán et al., 2007). Pollen records were expressed as the numbers of pollen grains per cubic metre of air (pollen grains $/m^3$). The method used 174 to calculate the MPS was the proposed by Nilsson & Persson (1981), which considers the 175 176 90% for the whole annual pollen amount (Fig.3).

177 2.4.Phenological analysis

The phenological phases studied were recorded according to the BBCH code (Biologische Bundesanstalt, Bundessortenamt, Chemische Industrie) (Meier, 1997). This is an internationally recognized standard in the agricultural sector and classifies plant growth phases according to a standardized system (Meire, 2001). From 15 specimens, but only on sunny days and with calm winds at noon, pollen shedding was mechanically tested from 10 branches at 1.5-2 m height that were touched or shaken, during four years (2016-2019) from April to July. Five specimens were 4 km from the pollen station; five specimens were 3.6 km distant; and five specimens were 3 km distant, while an average sampling frequency of 3-4
days was used. Specimens close to the pollen trap are shown in Fig. 1B. The plots for the
MPS and the phenological analysis is shown In Fig. 4.

The percentages of pollen shedding were recorded from beginning of flowering with 10% open flowers (BF, BBCH 61) up to full flowering (FF, BBCH 65, general flowering or full blossom) when at least approximately 50% of the flowers were open. This methodology has also been previously used in other works (Monroy-Colín *et al.*, 2018).

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2.5.HYSPLIT analysis

193 The air mass transport patterns above Badajoz were examined using daily 24-hours 194 backward dispersion analysis, following the methodology explained in previous works (De Weger et al., 2015). According to the hourly analysis (Fig. 5), the maximum peaks for 2017 195 and 2018 were observed from 13-15 hours (Fig. 5B and 5C), and it was the time set up for 196 the arrival of air masses. These plumes were calculated with the HYSPLIT model (Draxler 197 & Hess, 1998; Draxler & Rolph, 2014; Rolph, 2014). The Global Data Analysis System 198 199 (GDAS) data with 0.5 x 0.5-degree resolution were downloaded from the NOAA ARL (National Oceanic and Atmospheric Administration Air Resources Laboratory) FTP server. 200 201 The position for the particles in the layers of the atmosphere was studied, from 0 to 4 000 202 meters height (AGL), and the starting time of 15 hours for two consecutive days before each episode were calculated to trace the path that the air masses followed. They were during the 203 mismatches between the MPS period and the phenological phenophases from 2016 to 2019. 204 205 The results are displayed in Fig. 7.

206 2.6. Statistical analysis

After checking the normality by using the Kolmogorov-Smirnov test and due to the 207 negative results, that were obtained, non-parametric statistics was applied. The Spearman's 208 209 rank coefficient test was used to analyse the associations between pollen concentrations and 210 meteorological parameters (temperature, rain, relative humidity, wind frequency and wind speed). To determine what temperature recorded the most pollen, a graphical analysis was 211 performed that summed the daily pollen concentrations for each average temperature. The 212 statistical analysis was performed with the package R (R Core Team, 2018). The calculations 213 for the MPS and other parameters for the MPS were carried out by the package AeRobiology 214 215 (Rojo et al., 2019).

216 **3.** <u>Results</u>

217 In the entire city of Badajoz and the immediate surroundings, a total of 2,217 Olea europaea trees were counted (Fig. 1B). Most of the olive tree crops in the Iberian Peninsula 218 219 are located below 765 m in elevation and are concentrated mainly in the south (Fig. 2). Olive 220 tree flowering in Badajoz started the last week in April and finished the second week in June, with a maximum in the second week of May (Table 1 and Fig. 4). The flowering started two 221 222 weeks earlier in 2017 (Table 1 and Fig. 3). The average temperature in the two months before the flowers opened was higher in 2017 and 2019 (Table 1). 2017 and 2019 presented high 223 pollen concentrations (Table 2 and Fig. 3). In 2016, the phenological period ranged from 224 225 06/05-03/06, with a maximum on 20/05 (Table 1); pollen recorded outside this period represented the 19.83% of the total pollen. Maximum pollen concentrations were reached 226 227 only one day after relative to the maximum pollination phenophase (Fig. 4A). For 2017, the Phenological period ranged from 21/04-19/05 (Table 1), with a maximum on 12/05; pollen 228 records outside this period represented the 14.54% of the total pollen. Maximum pollen 229 230 concentrations were reached nine days before the maximum pollination phenophase (Fig. 4B). By contrast, in 2018 the Phenological period ranged from 11/05-01/06, with a maximum 231 on 18/05 (Table 1); pollen records outside this period represented the 36.71% of the total 232 233 pollen. The maximum pollen concentrations were reached five days later relative to the maximum pollination phenophase (Fig. 4C). In 2019, the Phenological period ranged from 234 22/04-21/05 (Table 1), with a maximum on 13/05; pollen records outside this period 235 represented the 14.83% of the total pollen. Maximum pollen concentrations were reached 236 237 two days later relative to the maximum pollination phenophase (Fig. 4D).

238 The phenological observations of Olea europaea (Table 1) indicate that pollination occurred for 26.5 days as the 4-year-average and was mostly within the period of the airborne 239 240 pollen records (Fig. 4). The pollen record dates outside the phenological observations 241 represented 19.29%. 4.35% of pollen was detected before the pollen season and 3.89% of pollen was detected after the pollen season. In 2016 and 2019, the maximum pollen 242 concentrations coincided with the maximum of the phenology periods, however, in 2017 and 243 2018 they did not. In 2017, the maximum concentration of pollen occurred nine days before 244 the maximum of the phenology period, while in 2018, it occurred five days later than the 245 246 phenological maximum (Table 1 and Fig. 4).

In 2016, the SPIn (Seasonal Pollen Integral, Galán et al., 2017) for the period studied was 247 2 606 pollen*day/m³ (Table 2). The daily peak was reached on 21/05 (451 pollen grains/m³ 248 (Fig. 4A), 18 days later than in 2017 (03/05), when a concentration of 1 994 pollen grains/ m^3 249 was reached (Table 2 and Fig. 4B). In 2017, the SPIn was 14 015 pollen*day/m³, while in 250 251 2018 it was 7 894 pollen*day/m³ (Table 2) and the daily peak value for pollen concentration reached 794 pollen grains/m³ (23/05) (Fig. 4C). 2019 was the year with the highest SPIn, 252 253 (considering the first semester data) with a total of 14 823 pollen*day/m³ (Table 2), and the daily peak reached 1 558 pollen grains/m³ (15/05) (Fig. 4D). 254

Table 2 show the SPIn for the studied period and the Spearman's correlations for 255 256 meteorology and daily concentrations regarding the MPS. Fig. 5 shows the hourly concentration patterns for 2016 (Fig. 5A), 2017 (Fig. 5B), 2018 (Fig. 5C) and 2019 (Fig. 5D). 257 258 During the four study years, the highest concentrations were found after noon, and we found a direct correlation with temperature increases and humidity decreases in the atmosphere. In 259 addition, the statistical analysis indicates a positive correlation with wind speed and negative 260 with wind frequency (Table 3). Fig. 6 shows the frequency of wind directions, and the highest 261 concentrations appeared when the winds were predominantly W and SW. 262

263 The pollen season of 2016 and 2019 were explained well because of local flowering 264 conditions (Fig. 4A and 4D), but not for 2017 and 2018 (Fig. 4 B and 4C). For these years, the years with the greatest difference between the maximum pollen concentrations and the 265 maximum phenological status, the backward dispersion calculation are displayed in Fig. 7. 266 They were the most likely to be due to LDT episodes. Thus, we can understand the effects of 267 long-distance transport that arrived from the SE zone of Andalusia (Fig. 7A and 7B), which 268 coincides with the region of greater *Olea* crop concentrations within the Iberian Peninsula 269 (Fig. 2). Moreover, we can observe that even for the episode occurred on 3rd May 2017, some 270 271 particles are coming from Portugal contained in air masses travelling for higher layers (>2 272 000 m AGL). For 2016, the difference between the maximum pollen concentration and the maximum phenology was one day (Tables 1 and 2). For 2018, the difference between the 273 274 maximum pollen concentration and the maximum phenology was five days (Tables 1 and 2). The air masses came mostly from the SW (Fig. 6C), but for the episodes of higher 275 concentrations out of the flowering season (they produced high peaks when the flowering 276 277 was decreasing), was found that the air masses were coming from distant places in Portugal

(7C, 7D, 7E and 7F). Finally, for 2019, at the time of the maximum pollen concentration, the
dominant wind direction was from the SW, the place where the most of trees are dispersed
within the city (Fig. 1 and 6D).

281 **4.** <u>Discussion</u>

282 The pollen season parameters were measured in the current work were similar to those reported by Aguilera et al. (2015b), Rojo & Pérez-Badia, (2015b) and Rodríguez-De la Cruz 283 284 et al. (2010), who reported maximum pollen peaks in the third week of May, but with lower values than reported in this work. However, the start date coincides with the reports for some 285 286 Mediterranean cities in Tunisia and southern Spain in the hottest years (2017 and 2019) 287 (Table 1), and these cities were those with average temperatures very similar to those found in Badajoz in those years. The dates for the beginning of the season during the coldest years 288 were coincident with the reported by Aguilera et al. (2015b) in some Italian cities with 289 290 continental climate. Latitude-induced microclimatic conditions determine the physiological responses of olive trees. In particular, the peak pollen emission date, which corresponds to 291 292 the day on which most of the tree canopy flowers are open, is influenced strongly by this factor (Aguilera et al. 2015b). The current findings confirmed that the air pollen counts were 293 294 determined by the flowering succession (Rojo & Pérez-Badia, 2015b). Olea pollen was 295 detected in the atmosphere of Badajoz from April to July. The flowering dates of the olive trees in the city of Badajoz coincided with that reported by Fornaciari et al. (2000), González 296 & Candau (1997) and Trigo et al., (2008) who reported that the flowering period in the 297 Mediterranean area for the genus Olea generally occurred between April and June. Moreover, 298 other authors in Spain have also reported that flowering took place at the end of May and in 299 the beginning of June (Rojo & Pérez-Badia, 2015a; Galán, et al. 2001a; González & Candau, 300 1997). 301

From a phenological point of view, **come early** the onset of the flowering period can be interpreted as a defence mechanism or an adaptive phenomenon of the olive tree physiology to the higher temperatures expected in the future and, above all, to the increases projected for the spring temperatures (Aguilera *et al.* 2015a). It has been observed that olive trees with constant maintenance and irrigation initiate flowering earlier and have longer flowering periods (Aguilera *et al.* 2013). Aguilera & Ruiz (2009) indicated that the accumulated temperature and precipitation during the months prior to the flowering period greatly favour the phenological development of the olive tree, and significantly affect the processes of
flower formation and the release of pollen grains into the atmosphere. Otherwise,
precipitation washes the atmosphere and causes a decrease and discontinuity in the presence
of pollen in the atmosphere.

313 It has been evidenced, using partial least-squares regression, that the reproductive phenology of olive trees in the Mediterranean area is regulated by meteorological parameters 314 315 that are related to the previous autumn and to both the winter and spring seasons, and, above all, by the temperatures (Aguilera et al., 2015a). Moreover, an increase in temperature before 316 the beginning and advance of flowering play a relevant role in the biological cycle 317 318 (Bonofiglio et al., 2008), and studies show that in the coldest years, the flowering period is shorter (Tedeschini, et al., 2006), while in the warmer years, the buds break earlier and in 319 this manner, flowering commences. González & Candau (1997) in a study in Sevilla, 320 indicated that the beginning of the main pollination period was related to the mean 321 temperature of the two preceding months and that pollination occurred when the average 322 323 temperature in both months was above 14°C. In contrast, pollination was delayed when the average temperature was lower. This agrees with the results of this study (Table 1), with the 324 325 opening of flowers occurring earlier in the hottest two years and with temperatures above 326 14°C (2017 and 2019). In addition to the release of pollen occurring gradually, this phenomenon could explain the differences in the beginning dates and durations of flowering 327 328 in 2017 and 2019 (Rojo & Pérez-Badia, 2015a; García-Mozo et al. 2008; Tedeschini, et al., 2006). Excessive temperatures negatively influenced several reproductive parameters, 329 including flower development, pollen-tube growth, pollen viability, and the pollination 330 process and, consequently, subsequent fertilization and productive yields (Aguilera et al. 331 2013). Other meteorological factors considered in relation to the phenology were humidity, 332 333 cumulative rainfall and cumulative solar radiation (Aguilera & Ruiz-Valenzuela, 2009). High 334 relative humidity sometimes can delay or even inhibit anther opening and pollen release (Yates & Sparks, 1993; Lisci et al., 1994). This could be another explanation for the observed 335 discrepancies, as shown in Fig. 4, in which it is possible to see the influence of rainfall. The 336 associated increase in the relative humidity provoked by rain during months with high 337 temperatures can provoke that pollen remains more time in the anthers, and brief showers 338 339 can also precipitate the abrupt deposition of pollen into the ground.

The values for the cumulative annual pollen mentioned in this study are only lower than 340 those for Córdoba, Jaen and Granada, as reported for Spanish cities that present amounts 341 above 22,000 pollen*day/m³ (Rojo *et al.* 2016). The *Olea* concentration values on the peak 342 days ranged from 414 pollen grains/m³ in 2016 to 1,994 pollen grains/m³ in 2017, and the 343 344 interannual variability in the different variables that define the pollen season is remarkable. Regarding the concentrations of Olea grains found on both the peak days and during the 345 346 pollen season, it should be noted that the olive tree is recognized as a species with alternation in flowering (Galera *et al.* 2018), which depends on vegetative and reproductive processes, 347 and occurs throughout a biennial cycle (Rallo & Cuevas, 2001); hence a part of the 348 349 interannual fluctuations is associated with this phenomenon (Galera et al. 2018). Olea pollen concentrations on both the peak days and in the pollen seasons (Table 2) show important 350 interannual variations, and those years with low concentrations (2016 and 2018) are followed 351 by years with high concentrations (2017 and 2019). This effect seems to be due to the 352 alternate bearing years of pollen production by this species (Rojo et al. 2015c) and coincides 353 354 with that reported by Galera et al. (2018) for the city of Cartagena, where the same effect was seen. 355

Considering some mismatches between phenology and pollen (Fig. 4) and for the 356 357 backward dispersion calculation analyses (Fig. 7), we suggest that for 2017 and 2018, the pollen outside the flowering season originated from distant sources (Fernández-Rodríguez et 358 al. 2014a). Specifically, for 2017 the peak for pollen was produced quite before the flowering 359 peak (9 days earlier), that could be due to pollen contributions from the SE of Badajoz, and 360 for 2018 the episodes showed air masses arriving from Portugal. Pollen transport from remote 361 sources is particularly relevant for species with small pollen size, such as the olive tree 362 363 (Hernandez-Ceballos et al. 2011). Other authors (Estrella et al., 2016) have also argued that 364 the discrepancy between pollen and phenology is due to LDT episodes. The pollen quantities that were recorded in 2017 before the flowering of olive trees in the area must have been 365 transported by winds from south and southeast of Spain, where the olive trees flowered 366 sooner due to higher temperatures and where earlier-flowering varieties are more widely 367 grown (García-Mozo et al. 2008). Specifically, winds from the south (May 3, 2017) could 368 have carried pollen from the olive tree groves in the Andalusian provinces of Jaen and 369 370 Granada which, along with Cordoba, have the largest olive tree growing areas in Spain.

Additionally, another pollen source in the west part of the territory could correspond to the 371 372 Alentejo region, which hosts nearly half of the olive tree groves of Portugal (Fernández-Rodríguez et al. 2014). It could be the reason for the episodes shown in Fig. 7 regarding 373 2018. Our results are consistent with the findings of Silva-Palacios et al. (2000) and 374 375 Fernández-Rodríguez et al. (2014), who demonstrated that, in Badajoz, there were significant correlations between the daily pollen concentrations of *Olea* and the amounts of wind hours 376 377 each day from the southeast, southwest and northwest quadrants. In this study, the HYSPLIT model has been proven to be an adequate tool to estimate the directions taken by air masses 378 379 and to identify the sources of pollen originating from long distances outside the pollen season 380 in the same region than for the current study (Maya-Manzano et al. 2016). Finally, we cannot discard the occurrence of some contribution from resuspension phenomena, especially in 381 2018. This fact has been proven to be important, above all in favourable conditions of high 382 temperature and absence of rain (Jato et al., 2006). In addition to the importance of 383 temperature, this study highlights the importance of wind, whether for long-distance 384 385 transportation or for nearby areas for those moments in that flowering peaks and pollen peaks are not coincident. This is probably related to the fact that the prevailing winds are from the 386 west (Fig. 5), where there are sources of olive tree pollen and that some of the highest 387 388 densities of olive tree groves in the Iberian Peninsula are found in the southeast (Fig. 3).

389

390 **5.** <u>Conclusions</u>

This type of studies facilitate better planning for the management of olive tree crops in 391 the region because they are useful to establish future predictions of the start of olive tree 392 flowering. LDT phenomena and delays in the flowering caused by environmental conditions 393 394 may lead to a non-exact match between the phenological and aerobiological peaks. At the 395 aerobiological level, LDT can be recognized by sudden appearances or increases (peaks) that 396 are sometimes coincident in different locations or by changes in the trends of atmospheric pollen dynamics, and LDT can be demonstrated using atmospheric models of air mass 397 dynamics. Backward dispersion calculations need to be combined with knowledge of the 398 geographical distributions and phenological characteristics of the pollen sources, as well as 399 with pollen concentration data of high temporal resolution, to clearly identify the origins of 400 401 pollen measured at a monitoring site. This information is extremely useful for predicting the amounts of airborne *Olea* pollen in the city of Badajoz and for other cities with similarconditions.

404

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Tables and figures

Table 1. Pollination phenology of *Olea europaea* (2016-2019). The onset, finish andmaximum dates are related to the flowering period.

	Onset Date	Finish Date	Maximum	Length	Tmean°C
				(days)	(two months before the flowers opened)
2016	06-May	01-Jun	20-May	26	12.8
2017	21-Apr	24-May	12-May	33	15.3
2018	11-May	08-Jun	18-May	28	13.1
2019	22-Apr	21-May	13-May	30	14.2

708 Table 2. SPIn (Seasonal Pollen Integral) values, mean pollen season characteristics and Spearman's rank correlation coefficients for

daily values daily values of *Olea europaea* 2016-2019 with rain (mm), Minimum (T_{min}), Maximum (T_{max}) and mean temperature (T_{mean}),

710 in °C, and Wind Speed (m s⁻¹).

* significance at the 95% level. ** significance at the 99% level.

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		Sease	Mean Pollen Season (MPS)					
	pollen*day m ⁻³	Spearman's	s rank coeffic	ient rank	Length	Maximum pollen concentration		
		Rain	T_{min}	T _{max}	T _{mean}	Wind speed	-	
2016	2 606	0.11	-0.04	_0.19	-0.12	-0.11	14/05 to 22/06	21/05
2010	2,000	0.11	-0.04	-0.19	-0.12	-0.11	(40 days)	(451 pollen grains/m ³)
2017	14 015	-0.22	0.01	0.09	0.1	-0.01	27/04 to 25/05	03/05
2017	14,015	-0.22	0.01	0.07	0.1	-0.01	(29 days)	(1,994 pollen grains/m ³)
2018	7 894	-0.04	-0.15	0.08	0.01	-0.09	17/05 to 23/06	23/05
2018	7,094	-0.04	-0.15	0.08	0.01	-0.09	(38 days)	(794 pollen grains/m ³)
2019	14,823	0.12	-0.07	-0.22	-0.2	-0.28	02/05 to 01/06 (31 days)	15/05 (1,558 pollen grains/m ³)

- Table 3. Spearman's rank correlation coefficients between hourly values of *Olea europaea*
- 715 2016-2019 during their MPS and hourly meteorological parameters.
- * significance at the 95% level. ** significance at the 99% level.

	Hourly values with the highest pollen	Hourly values with the lowest pollen	Statistically significant correlation				
	concentration	concentration	Relative	Wind	Temperature	Wind	
			humidity	speed	Temperature	frecuency	
2016	10:00-15:00	22:00-09:00	-0.64**	-0.74**	0.59**	-0.64**	
2017	11:00-18:00	23:00-07:00	-0.73**	0.89**	0.73**	-0.76**	
2018	13:00-18:00	22:00-09:00	-0.84	0.42*	0.86*	-0.84**	
2019	10:00-17:00	23:00-08:00	-0.82**	0.27	0.43*	-0.59**	

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- Fig. 1 A. Pollen trap and portable weather station (WS-GP1 Delta-T). B. Geolocation of *Olea*trees. Spore trap (red dot). Olive trees (blue dots). Specimens whose phenology was studied
 (yellow dots)





Fig. 2. Mapping sources of olive pollen of the Iberian Peninsula with olive groves (blackcolour) from CLC and elevation data in Badajoz (BA).





Fig. 3. Pollen Season of *Olea europaea* 2016-2019.



Fig. 4. Daily airborne olive pollen concentration (blue lines), mean temperature (orange lines), rain (black bars), MPS dates for start and
end (blue bars) and phenology (red dots). 2016 (A), 2017 (B), 2018 (C) and 2019 (D).







Fig. 5. Hourly data for Olea pollen concentrations in 2016 (A), 2017 (B), 2018 (C) and 2019

Fig. 6. Wind direction frequency during pollen season of *Olea europaea* in 2016 (A), 2017
(B), 2018 (C) and 2019 (D).



Fig. 7. HYSPLIT model calculation for the 24-hours back-distribution of particles released in Badajoz for the mismatches between pollen peaks and flowering peaks in 2017 (May 3rd A and B) and 2018 (May 30th C and D, and June 6th E and F).



