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Pollen couriers across the Mediterranean: the case of migrating warblers

Jacopo G. Cecere[1,*], Laura Cornara[2], Serena Mezzetta[2], Andrea Ferri[3], Fernando Spina[3] & Luigi Boitani[1]

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At stopovers sites in the Sahara and on Mediterranean islands, European warblers may feed on nectar. As a consequence of this behaviour the pollen of exploited plants often sticks on the birds' bill or plumage. We analyzed pollen samples carried by migrating warblers staging at Ventotene Island (Italy), a stopover site located in the Central Mediterranean and reached by large numbers of birds after a direct flight from North Africa. We discuss the possible role of migrants as pollen vectors. A total of 38 plant taxa were recorded in the pollen samples collected on 147 migrants. Pollen of *Brassica* sp., *Ferula communis*, *Citrus* sp. and *Eucalyptus* sp. were most commonly found. The nectar of the first two plants is largely exploited by warblers on Ventotene and the presence of their pollen on plumage is principally due to nectar feeding *in situ*. On the contrary, *Citrus* sp. and *Eucalyptus* sp., which are scarce and absent on the island respectively, were mainly collected on dried and agglutinated samples and their presence is likely to be due to nectar feeding at other sites than Ventotene. The other 34 plant taxa were recorded with low frequencies. Overall, the data suggest that nectar feeding migrants could play a role as pollen vectors only for *Citrus* sp., *Eucalyptus* sp., *F. communis* and *Brassica* sp. However, the other less common plant taxa could also be useful markers of migration routes. We found species-specific relationships between bird species and the plants used for nectar feeding: short-distance migrants (mainly Eurasian Blackcap *Sylvia atricapilla* and Subalpine Warbler *S. cantillans*) land on Ventotene when *Brassica* sp. flowers are more abundant and they mainly carry the pollen of this species. On the contrary, long-distance migrants (Garden Warbler *S. borin* and Common Whitethroat *S. communis*) principally transport the pollen of *F. communis*, whose flowering period takes place at the same time as their staging period on the island. The study underlines the tendency of warblers towards nectar feeding during migration.

Key words: *Brassica*, *Citrus*, *Eucalyptus*, nectar feeding, nectarivores, pollen vectors, *Sylvia*

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Every year more than 2 billion passerines move between Africa and Europe (Hahn *et al.* 2009). Several of these species, mainly warblers, are occasional nectarivores, which land at stopover sites with pollen grains stuck on their body (Cecere *et al.* 2010). Nectar feeding by warblers is well described at a stopover site located in the Sahara desert (Salewski *et al.* 2006) but also at Ventotene, an Italian island reached by birds after the crossing of the Mediterranean Sea during spring migration (Schwilch *et al.* 2001). Nine species of passerines are recorded as nectar feeders on Ventotene Island, six of which are trans-Saharan migrants. These

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birds use at least six plants belonging to different families to forage on nectar: *Ferula communis*, *Brassica* sp., *Lavatera arborea*, *Prunus avium* and *Pittosphorum tobira* and probably *Malva sylvestris* (Cecere *et al.* 2011). As a consequence of nectar feeding, pollen grains can remain stuck to the bill and the forehead of the birds (Ash 1959, Ash *et al.* 1961, Laursen *et al.* 1997, Schwilch *et al.* 2001, Ortega-Olivencia *et al.* 2005, Salewski *et al.* 2006). In Denmark, Laursen *et al.* (1997) found pollen grains belonging to at least nine plant taxa on the bill of some migrating warblers. In the same study, the authors found pollen belonging to plants that are abundant in the Mediterranean region and absent in Denmark, and they suggested that the pollen found on birds can be used as a marker for migratory routes.

Similarly to exotic birds, European songbirds seem to be able to pollinate plants by carrying pollen grains on their plumage and bill. This has been shown for Blue Tits *Cyanistes caeruleus*, which can pollinate an exotic plant, *Fritillaria imperialis*, in the United Kingdom (Búrquez 1989), but also for Sardinian Warbler *Sylvia melanocephala*, Eurasian Blackcap *S. atricapilla* and Chiffchaff *Phylloscopus collybita*, which regularly pollinate the European native plant *Anagyris foetida* in Southern Spain (Ortega-Olivencia *et al.* 2005).

Despite several reports about pollen transport by Old World songbirds, an in-depth analysis of pollen species carried by migrants landing in Europe and coming directly from Africa is missing. This kind of analysis is necessary to assess which plants are used for nectar consumption at stopover sites by songbirds crossing the Mediterranean Basin. Furthermore, it can show specific relationships between bird and plant species. Overall, this information can add to the knowledge of the ecology of European migrants and inform habitat management at stopover sites. Lastly, these studies could be able to assess the role of European songbirds in plant pollination.

In this paper we analyze pollen grains found on 147 birds belonging to eight species landing at a stopover site located in the Central Mediterranean during spring migration, and discuss the possible role of migrants in the dispersion of pollen. Moreover, we analyze the flowering patterns of the two plants most frequently used by songbirds on Ventotene Island and the staging patterns of occasional nectarivore migrants at the stopover sites (Cecere *et al.* 2010), in order to assess whether there is a relationship between flower availability and pollen grains found on bird species, thus between bird species and the plants they feed on.

METHODS

The study was carried out in spring at Ventotene Island during the years 2006–2009. The island (1.3 km^2) is 50 km off the Tyrrhenian coast of Italy; it is inhabited by a small community (450 people) and covered by several small fields characterized by crop rotation (cultivated and set aside). Since 1998, the Italian Ringing Centre (CNI-ISPRA) has carried out the "Progetto Piccole Isole – Small Islands Project" at a large network of sites, including Ventotene, in order to investigate stopover strategies adopted by songbirds during spring migration (Spina *et al.* 1993). In this study, continuous and standardised ringing activity was carried out from 1 April to 31 May. A total length of 282 m of mist-net was checked for the presence of birds every hour and every day, from one hour before sunrise till one hour after sunset; nets were not operated in bad weather conditions (strong wind or heavy rain).

Seasonal flowering patterns and presence of migrants

Seasonal flowering patterns were obtained for the two plants most used by passerines on Ventotene, following Cecere *et al.* (2010): *F. communis* (Apiaceae) and *Brassica* sp. (Brassicaceae). Anzalone & Caputo (1976) referred to the presence of *Brassica oleracea* L. ssp. *robertiana* (Gay) Rouy et Fouc. and *B. fruticulosa* Cyr. ssp. *fruticulosa* on the island. However, *Brassica* is an important genus in agriculture, with a wide range of crops and it is likely that hybridisation may have occurred in the past few decades. Taking into account this possibility, and the well known taxonomic uncertainty of this genus (Gladis & Hammer 2001), we refer to this species as *Brassica* sp.

The census of blooming *Brassica*. sp. and *F. communis* was carried out within 10 plot areas in 2006 and 2007. All flowers on plants inside 5 plots were counted every five days, from 1 April to 31 May. Plots had an area of 2 m^2, were placed at least 200 m from each other on land with the following exposures: SE, E, NE, N, and W. In the second year, each plot was moved a few meters in order to sample different plants. For *Brassica* sp. all open flowers were directly counted. In the case of *F. communis*, the percentage of open flowers per each inflorescence was estimated. Generally, the branches of *F. communis* have inflorescences of two sizes, which were categorised as big or small. After each count, we computed the flowering index of *F. communis* as follows:

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$$((\sum_{x=1}^{N} Fop_x / Ftb_x) + (\sum_{x=1}^{N} Fop_y / Fts_y)/2)/1000,$$

where *FoP* = open flowers; *Ftb* = total flowers of big inflorescence; *Fts* = total flowers of small inflorescence; *N* = total number of big inflorescences; *M* = total number of small inflorescences. This formula allowed us to obtain an index of abundance of blooming *F. communis* by the estimated percentage of open flowers for each inflorescence present within the plot areas. The index was used because a count of each single little flower was not possible without dissecting the inflorescence.

The migrating patterns of the four most common migrants landing on Ventotene, which regularly feed on nectar, viz. Eurasian Blackcap, Subalpine Warbler *S. cantillans*, Garden Warbler *S. borin* and Common Whitethroat *S. communis*, (Schwilch *et al.* 2001, Cecere *et al.* 2010) were estimated by summing up all captures made every five days in 2006 and 2007. Because data were not always normally distributed (Shapiro–Wilk test), trends in 2006 and 2007 were compared by Kolmogorov–Smirnov test, both for the four migrating species and for blooming *Brassica* sp. and *F. communis*.

Pollen collection and frequency classes

The collection of pollen from birds (Fig. 1) was carried out from 10 April to 20 May 2008, a period encompassing the flowering peaks of *F. communis* and *Brassica* sp. The survey was carried out along with ringing activities. Pollen was collected from the beak, forehead or breast feathers of trapped birds using a short plastic stick with a small amount of basic fuchsin gelatin at one end (200200, Lanzoni s.r.l., Bologna, Italy). This method allowed us to collect pollen samples without harming the birds. Each stick was then carefully introduced into a test tube and conserved at room temperature. Each pollen lump on the plumage of birds was classified as fresh or dried. In fresh pollen, grains are still visible as they are on the stigma (Fig. 1C). On the contrary, dried pollen is identifiable as it is agglutinated, generally thicker than 1 mm, and encrusted on the feathers (Fig. 1D). In the laboratory, the stained jelly containing pollen samples was heated at 40°C, allowing us to make semi-permanent microscope slides of pollen. A total of 147 slides were prepared, each corresponding to a single trapped bird. Slides were observed using a light microscope and pollen grains were counted by reading 3 randomly-chosen, longitudinal bands, each 1/3 mm in diameter, by using 40× objective and



Figure 1. A) Garden Warbler feeding on the nectar of *Ferula communis*. B) Garden Warbler on *F. communis* with breast dirty with nectar and pollen (photos by Marzia Mirabile). C) Garden Warbler showing fresh pollen on the bill and forehead. D) Garden Warbler with dried pollen on the bill and forehead (photos by Jacopo G. Cecere).

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10× ocular lenses (Spieksma 1983). Pollen was identified at the species level, or in some cases at the genus or family levels. The taxonomic identification was based on the palynotheca of the "Polo Botanico Hanbury", University of Genova, pollen reference slides prepared from the anthers of flowers collected in the sampling area, or literature reports (Faegri 1993, Ricciardelli D'Albore *et al.* 1978, Punt & Clarke 1981, Valdés *et al.* 1987, Reille 1992, Bucher *et al.* 2004).

As suggested by Louveaux *et al.* (1978), we used the following terms for defining pollen frequency classes in each slide: 'predominant', when a pollen species showed a frequency higher than 45% of all recorded pollen grains in the slide; 'secondary' (frequency = 16–45%); 'important minor' (frequency = 3–15%); 'minor' (frequency < 3%). The pattern of predominant plant species collected in dry samples was compared with that in fresh samples by a Chi-square test. The same test was also used to compare predominant plant species recorded on the body of short and long distance migrants.

RESULTS

Seasonal flowering patterns and presence of migrants

The flowering pattern of *Brassica* sp. did not differ between 2006 and 2007 (Kolmogorov–Smirnov, $Z = 0.4$, $P = 1$), despite some slight differences: during 2006, *Brassica* mainly flowered between the first five days of April and the first days of May, with a peak between 11 and 15 April; in 2007, the flowering period had a shift, starting after 10 April and ending at end of May, and the peak was longer than in 2006, lasting from 26 April till 5 May (Fig. 2). *F. communis* showed a similar pattern in the two years (Kolmogorov–Smirnov, $Z = 0.2$, $P = 1$), with a peak between 26 and 30 April; after 20 May, open flowers were rare on both plants (Fig. 2).

The four bird species showed similar staging patterns on Ventotene in the two study years (Kolmogorov–Smirnov, Eurasian Blackcap: $Z = 0.82$, $P = 0.52$; Subalpine Warbler: $Z = 0.61$, $P = 0.85$; Garden Warbler: $Z = 0.41$, $P = 1$; Common Whitethroats: $Z = 061$; $P = 0.85$), despite different annual ringing totals. Eurasian Blackcap, as well as Subalpine Warbler, migrate mainly during the first half of April and become scarce after 20 April; on the contrary, Garden Warblers are numerous during the second half of May, while Common Whitethroats are scarce in the first half of April and show a peak between April and May (Fig. 3).

Pollen collection

A total of 147 pollen samples (95 fresh and 52 dried) were collected on as many birds. With regards to fresh pollen, an average number of 235.3 ± 2.3 ($n = 82$) pollen grains/sample were recorded on a fraction of 86.3% of samples, 50–200 pollen grains/sample (100 ± 16.6, $n = 8$) on 8.4% of samples, while less than 50 pollen grains/sample (8.4 ± 4.1, $n = 5$) were recorded on 5.26% of samples. As for dry samples, over 200 pollen grains/sample (245.1 ± 7.8, $n = 17$) were recorded on a fraction of 32.7% of samples, 50–200 pollen grains/sample (99.4 ± 8.6, $n = 18$) on 34.6% of samples, and less than 50 (16.3 ± 3.01, $n = 17$) on 32.7% of samples. Frequency classes were not defined for samples with less than 50 pollen grains. As a consequence, the total number of classified samples was 125 (90 fresh and 35 dried). On the whole, pollen grains from 38 plant taxa were recorded in all samples, of which 7 taxa are certainly not present on Ventotene, while 3 taxa are very rare (Anzalone & Caputo 1976; pers. obs.). The lists of plant species recorded in fresh and dried samples are reported in Table 1 A and B, respectively.

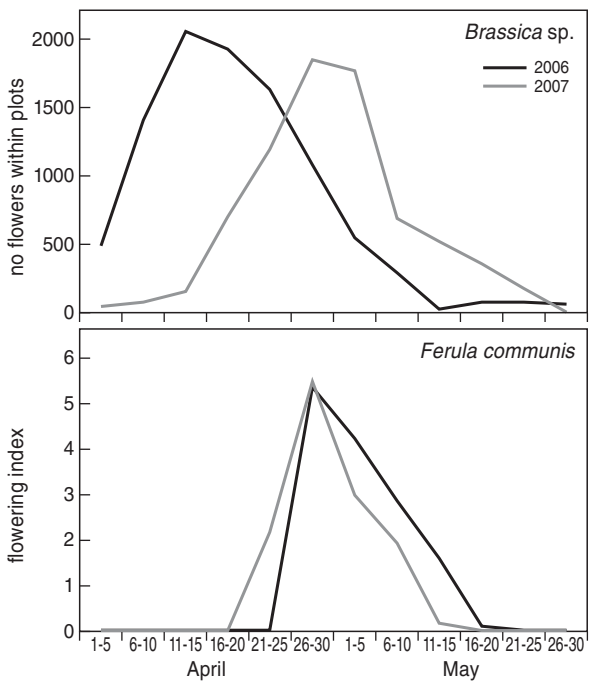


Figure 2. Flowering of *Brassica* sp. and *Ferula communis* recorded from 1 April to 30 May, in 2006 and in 2007. For *Brassica*, the trend was obtained by counting flowers in 10 plot areas, while for *Ferula communis*, the trend was obtained by a flowering index calculated after each control (for details see Methods).

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A total of 26 plant taxa were recorded in fresh samples (Table 1A), among which *Brassica* sp. (Fig. 4A) was the most frequent. This taxon was found in 72.2% of fresh samples and, among these, it was the predominant pollen species in 75.4% of cases. *F. communis* (Fig. 4B) was present in 51.58% of fresh samples and, among these samples it was the predominant pollen species in 81.63% of cases. Pollen grains from another 24 plant taxa were recorded with lower frequencies in fresh samples, and their presence was generally classified as 'important minor pollen' or 'minor pollen', except for *Malvaceae* (Fig. 4C), which represented the 'predominant pollen' in one fresh sample (Table 1A).

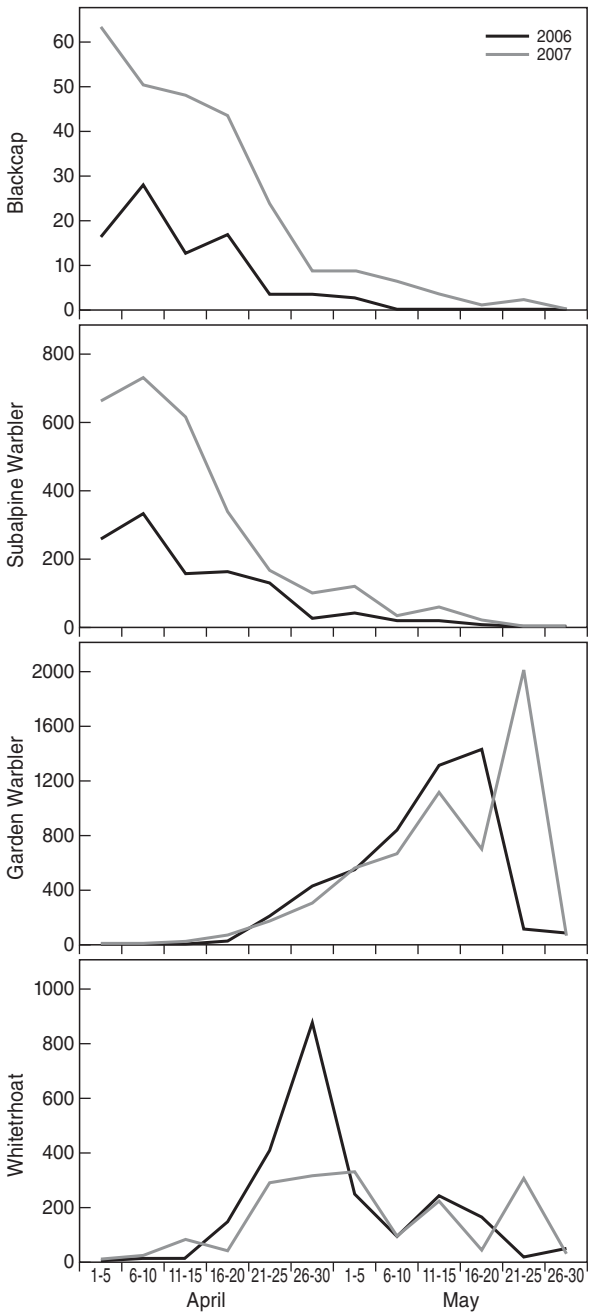


Figure 3. Estimated trends of landings of four migrants on Ventotene island, derived from the ringing activity carried out from 1 April to 30 May, in 2006 and in 2007.

Plant species that contributed to the predominant pollen in dry samples differed from those in fresh samples ($\chi^2 = 73.35$, $df = 3$, $P < 0.0001$); *Malvaceae*, *Cyperaceae* and *Fraxinus* sp. were excluded from the analysis since each of them was present as predominant pollen in only one sample. A total of 32 plant taxa were recorded in dry pollen samples (Table 1B) and among these, *F. communis* was the most frequent species. It was present in 23.2% of dry samples, and among these, it was the predominant pollen in 40.9% of cases. *Citrus* sp. (Fig. 4D) was the second most frequent pollen taxon, present in 21.05% of dry samples and, among these, it was the predominant pollen in 65% of cases. *Brassica* sp. was the third most frequent pollen taxon, present in 17.9% of dry samples and, among these it was the predominant pollen in 11.7% of cases. The fourth most abundant pollen taxon was *Eucalyptus* sp. (Fig. 4E), present in 11.6% of dry samples, and among these, it was the predominant pollen in 72.7% of cases. The other 28 plant species showed lower frequencies, being classified in most cases as important minor pollen or minor pollen (Table 1B).

Plant species recorded as predominant among fresh samples differed between short and long-distance migrants ($\chi^2 = 77.35$, $df = 1$, $P < 0.0001$). All Eurasian Blackcaps sampled with fresh pollen ($n = 7$) and 97% of Subalpine Warblers ($n = 37$) had *Brassica* sp. as predominant pollen. On the contrary, the two long-distance migrants carried mainly pollen of *F. communis*: 94% of Garden Warblers ($n = 35$) and 100% of Common Whitethroats ($n = 7$). *Malvaceae* was excluded from the analysis since it was present only in one sample.

Unfortunately, data and sample size did not allow us to perform a Chi-square test for dry samples. Nevertheless, differences between short and long-distance migrants in recorded predominant plants appeared to be present among dry samples too. *Citrus* sp. was the most common taxa as the predominant plant among dry samples collected on the two short-distance migrants: 60% of Subalpine Warblers ($n = 10$) and 50% of Eurasian Blackcaps ($n = 10$), while it was absent among dry samples collected on the plumages of long-distance migrants. On the contrary, *F. communis*

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Table 1. Occurrence of plant species per abundance class recorded among samples of pollen collected on the plumage and bill of migrating songbirds staging at Ventotene Island (Italy) during spring 2008. A) In fresh samples, and B) in dried samples. Pollination mechanisms: A = anemophilous; Z = zoophilous; M = mixed anemophilou– zoophilous (Oberdorfer 1949, Dommée *et al.* 1999, Horlock *et al.* 2000, Smeins 2003, Escudero *et al.* 2008).

Plant	Presence on total slides (%)	Predominant (%)	Secondary (%)	Important minor (%)	Minor (%)	Pollination mechanisms
A) Fresh samples						
Brassica sp.	72	75	2	2	22	Z
Ferula communis	52	82	0	2	16	Z
Gramineae	8	0	0	0	100	A
Fraxinus sp. **	8	0	0	13	88	M
Urticaceae	7	0	0	14	86	A
Quercus ilex +	5	0	0	40	60	A
Quercus sp. (no *Q. ilex*) **	4	0	0	0	100	A
Citrus sp. +	4	0	0	25	75	Z
Olea europea *	4	0	0	25	75	A
Pelargonium sp.	3	0	0	0	100	Z
Ostrya sp. **	3	0	0	0	100	A
Compositae	3	0	0	0	100	M
Euphorbia dendroides	3	0	0	0	100	Z
Pistacia sp.	2	0	0	50	50	A
Malus sp. +	2	0	0	0	100	Z
Tamarix sp.	2	0	0	0	100	A
Malvaceae	1	100	0	0	0	Z
Carpobrotus sp.	1	0	0	0	100	Z
Cupressaceae	1	0	0	0	100	A
Acacia sp. **	1	0	0	0	100	Z
Eucalyptus sp. **	1	0	0	0	100	Z
Prunus sp.	1	0	0	0	100	Z
Castanea sativa *	1	0	0	0	100	M
Rosaceae	1	0	0	0	100	Z
Dipsacaceae	1	0	0	0	100	Z
Pinaceae *	1	0	0	0	100	A
B) Dried samples						
Ferula communis	23	41	5	27	27	Z
Citrus sp. +	21	65	10	10	15	Z
Brassica sp.	18	12	35	24	29	Z
Eucalyptus sp. **	12	73	0	9	18	Z
Urticaceae	12	0	0	9	91	A
Fraxinus sp. **	11	10	20	10	60	M
Quercus sp. (no *Q. ilex*) **	8	0	13	13	75	A
Gramineae	8	0	0	0	100	A
Pinaceae *	5	0	0	20	80	A
Cupressaceae	4	0	0	0	100	A
Rosaceae	3	0	0	0	100	Z
Compositae	3	0	0	0	100	M
Ericaceae	3	0	0	0	100	M
Ostrya sp. **	3	0	0	33	67	A
Chenopodiaceae	2	0	0	0	100	A
Plantago sp.	2	0	0	0	100	A
Pistacia sp.	2	0	0	0	100	A
Prunus sp.	2	0	50	0	50	Z
Tamarix sp.	2	0	0	0	100	A
Euphorbia dendroides	1	0	0	0	100	Z
Malvaceae	1	0	0	0	100	Z
Rumex sp.	1	0	0	0	100	A
Labiatae	1	0	0	100	0	Z
Malus sp. +	1	0	0	100	0	Z
Liliaceae	1	0	0	0	100	Z
Cistaceae	1	0	0	0	100	Z
Betula sp. **	1	0	0	0	100	A
Pelargonium sp.	1	0	0	0	100	Z
Robinia sp. +	1	0	0	0	100	Z
Celtis sp. **	1	0	0	0	100	A
Cyperaceae	1	100	0	0	0	A
Olea europea *	1	0	0	0	100	A

** Absent on Ventotene; * Very rare; + Scarce (Anzalone & Caputo 1976; pers. obs.).

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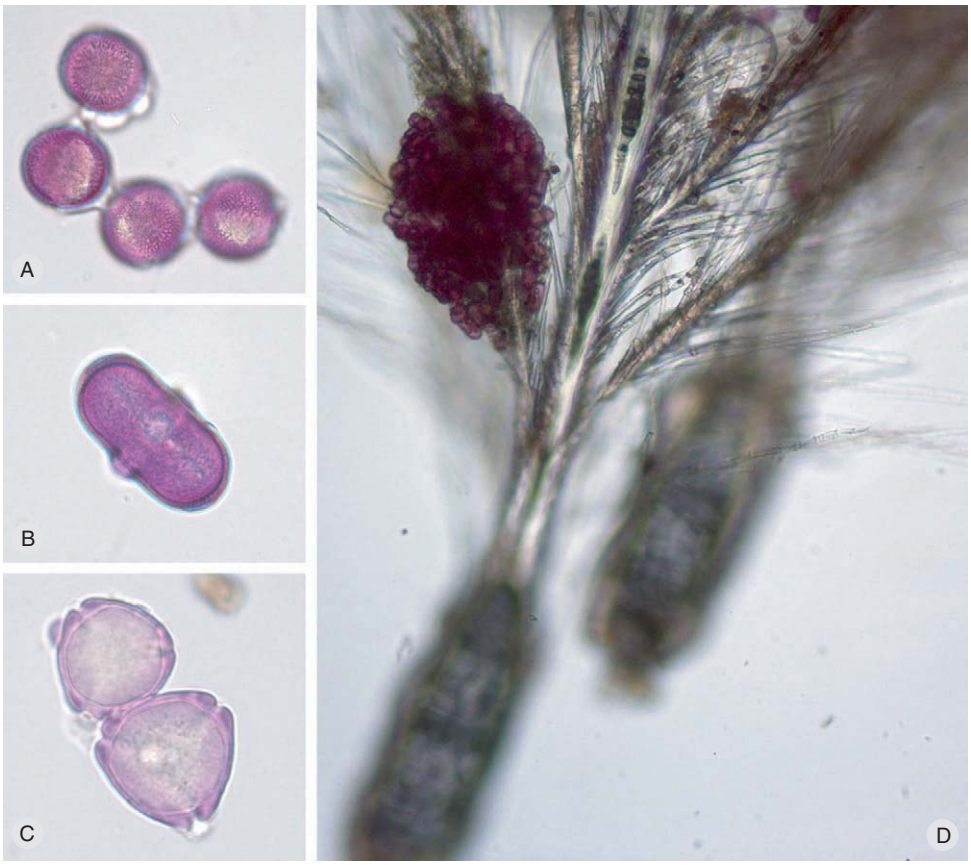


Figure 4. Pollen grains of A) *Brassica* sp., 63×, B) *Ferula communis*, equatorial view, 63×, C) *Citrus* sp., 63×. D) Aggregate of pollens stuck to a feather, 10×.

was recorded only on Garden Warbler, (60%; $n = 10$) and on the two sampled Common Whitethroats. Pollen grains of *Eucalyptus* sp. were found both on short and distance migrants (40% of Subalpines and 20% of Eurasian Blackcaps and Garden Warblers).

Pollen percentages were not calculated for Common Whitethroat with dry pollen or for the other species, Sardinian Warbler, Chiffchaff, Wood Warbler *P. sibilatrix* and Lesser Whitethroat *S. curruca*, due to small sample sizes (sample size of each species < 4).

DISCUSSION

In this study we show that pollen grains of a large array of plant species were carried every spring across the Mediterranean Region by migrating warblers. On the plumages of 147 birds staging at the stopover site of Ventotene Island, we found pollen grains belonging to 38 different plant taxa, seven of which are absent from the island (Table 1). This can only be explained by nectar feeding of warblers at previous African stopover sites (e.g. see Salewski *et al.* 2006) and on Mediterranean islands (Cecere *et al.* 2010). In addition, many other pollen species stick accidentally onto the plumage of birds, such as pollen from anemophilous and nectar-less plants. The nine plant taxa recorded on the plumage of migrants trapped in Denmark had also been found on migrants landing on Ventotene, with the exception of *Cannabis* sp. (Laursen *et al.* 1997).

In our study, *Brassica* sp. and *F. communis* were by far the most common species in the fresh pollen samples, which is consistent with the importance of these plant species for nectar-feeding songbirds on Ventotene (Schwilch *et al.* 2001, Cecere *et al.* 2010). However, these two plant species showed a different distribution among bird species. Almost all fresh pollen samples collected on Subalpine Warblers and Eurasian Blackcaps contained *Brassica* sp. as the predominant pollen, while almost all Garden Warblers and Common Whitethroats carried *F. communis*. The first two bird species are short-distance migrants staging at Ventotene

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earlier than Sub-saharan migrants, coinciding with the flowering peak of *Brassica* sp. (Fig. 2 and 3). In contrast, Garden Warbler and Common Whitethroat are long-distance migrants passing later in the season, when the number of flowers of *Brassica* sp. starts decreasing, while the flowers of *F. communis* are abundant (Fig. 2 and 3). This suggests that the pollen of fresh samples may have originated from nectar feeding *in situ*.

The remaining 24 plant taxa found in the fresh samples were only recorded with low frequencies, and a good proportion (38.5%) of the total taxa found in fresh samples belong to anemophilous plants (Table 1A). In these cases, the presence of pollen on plumage does not originate from nectar feeding, but rather from the wind or through simple contamination when the birds perch on such plants.

The pollen found in dry and agglutinated samples was likely to derive mainly from sites visited by migrants before their staging at Ventotene (Cecere *et al.* 2010). Most spring migrants spend less than two days on Ventotene island (Goymann *et al.* 2010; Tenan and Spina, unpubl. data) and this period is likely too short for pollen to dry. The pollen composition of dry and fresh samples were quite different: *Eucalyptus* sp., *Citrus* sp. and *F. communis*, were mainly present in dried samples as predominant pollen, while *Brassica* sp. was also common, but mainly present as secondary pollen. The other 28 plant taxa recorded in the dry samples showed very low frequencies and, like in fresh samples, a good proportion (46.87%) of the total taxa found in dry samples belonged to anemophilous plants (Table 1B).

F. communis and *Brassica* sp. are abundant on Ventotene but also at many other sites within the Mediterranean; *Citrus* sp. and *Eucalyptus* sp. are cultivated in the whole Region, but are scarce and absent, respectively, on Ventotene. Pollen grains of *Citrus* sp. were recorded on migrating *Sylvia* warblers trapped in the United Kingdom and in France (Ash 1959, Ash *et al.* 1961). *Eucalyptus* and *Citrus* were the genera most abundant among the pollen samples collected on warblers trapped in Denmark by Laursen *et al.* (1997). Based on our results, the pollen grains of *Citrus* sp. and *Eucalyptus* sp. were mainly carried by the short-distance migrants Eurasian Blackcap and Subalpine Warbler, whereas Garden Warbler, which is a long-distance migrant wintering in tropical and equatorial areas, was mainly found carrying *F. communis*.

The difference in species composition between the pollen carried by long- vs. short-distance migrants may be due to the different migration phenologies across the Mediterranean Region, which coincide with different flower availability, as occurs at Ventotene for *F. communis* and *Brassica* sp. In addition, different migration routes and patterns may have a role. Short and long-distance migrants could use different stopover sites, characterized by dissimilar vegetations; thus they may come into contact with different nectar resources, carrying, consequently, different pollen species on their plumages.

We hypothesize here that migrating songbirds, and in particular short-distance migrants, can play a role as pollen vectors of *Citrus* and *Eucalyptus* species through the Mediterranean region, especially when considering that some migrants land on Ventotene coming directly from North Africa (Pilastro *et al.* 1995). But in the case of dried samples, we have also to consider that pollen might not be viable for pollination.

Migrants could also have a role as pollen vectors in the interbreeding of *Brassica* sp. growing at Ventotene, where the plant is largely exploited by birds, or across the Mediterranean. The pollen grains of *Brassica* sp. were common on bird plumages, principally on the bill, as for *Citrus* sp. and *Eucalyptus* sp., and the plant is characterized by large flowers. On the contrary, it is harder to hypothesize a role of songbirds as pollen vectors for *F. communis*, since it is characterized by small flowers and its pollen is mainly found stuck on breast feathers. We cannot exclude that when a bird is nectar-feeding at a flower, the breast feathers can accidentally pollinate other flowers of the same inflorescence. Contrary to *Citrus* and *Eucalyptus* species, pollen of *Brassica* sp. and *F. communis* were not found on the body of migrants trapped at other Mediterranean sites. However, this may depend on a lack of data due to the very few studies on this topic in Europe, and future studies are necessary to assess this idea.

For other plant taxa, migrating warblers seem not to play a relevant role in pollen dispersal. The pollen of these plants was scarce on bird plumages and mostly derived from anemophilous plants. However, as suggested by Laursen *et al.* (1997), less frequent pollen taxa can be used as markers for the distribution of stopover areas visited before landing at the trapping site.

A recent study shows that warblers gain a physiological benefit from nectar consumption: nectar uptake, in fact, led to an increase in the plasma blood glucose concentrations of migrants (Cecere *et al.* 2011). With the present study we showed that as a consequence of nectar feeding, European warblers carried large amounts of pollen grains across the Mediterranean, suggesting a possible role of songbirds as vectors for

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pollination. A possible mutualistic relationship might thus occur between *Sylvia* warblers and some plant species, as for *Brassica* sp., *Citrus* sp. and *Eucalyptus* sp. Moreover, the large number of plants recorded in pollen samples marks the tendency of migrating songbirds to use nectar as a resource during spring migration. In particular, in urban and semi-urban habitats, generally less favourable to migrants, the presence of plants that flower between March and May can be an aid for birds to restore themselves during migration.

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SAMENVATTING

Sommige Europese zangvogels eten nectar tijdens de trek door de Sahara of op eilanden in de Middellandse Zee. Daarbij blijft vaak stuifmaal achter op de snavel of het verenkleed. Het onderhavige onderzoek richtte zich op stuifmeel dat werd verzameld van zangvogels die op het Italiaanse eiland Ventotene in de Middellandse Zee pleisterden na vanuit Noord-Afrika overgestoken te zijn. Bij 147 trekvogels werden 38 verschillende planten

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(soorten, geslachten of families) vastgesteld. Het stuifmeel was vooral afkomstig van Kool *Brassica* spec., Reuzenvenkel *Ferula communis*, Citrusvruchten *Citrus* spec. and Eucalyptus *Eucalyptus* spec. De twee eerstgenoemde planten komen veel voor op het eiland, zodat het stuifmeel afkomstig kan zijn van plaatselijke planten. Citrus is een schaars voorkomende plant, Eucalyptus komt niet op het eiland voor, zodat het stuifmeel van elders afkomstig moet zijn. Zangvogels zouden een belangrijke rol als verspreider van stuifmeel van de vier genoemde planten kunnen spelen. De overige 34 planten werden zo weinig vastgesteld dat deze rol minimaal moet zijn. Wel kunnen de vastgestelde soorten een aanwijzing geven van de trekroutes van de zangvogels. Trekvogels droegen het stuifmeel van planten die het meest talrijk bloeiden op het moment van doortrek. Korteafstandstrekkers (Zwartkop *Sylvia atricapilla* en Baardgrasmus *S. cantillans*) droegen vooral stuifmeel van Kool, langeafstandstrekkers (Tuinfluiter *S. borin* and Grasmus *S. communis*) stuifmeel van Reuzenvenkel. (ND)

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