

Supporting Information for

Frugivore-mediated seed dispersal in fragmented landscapes: Compositional and functional turnover from forest to matrix

Juan P. González-Varo, Jörg Albrecht, Juan M. Arroyo, Rafael S. Bueno, Tamara Burgos, Gema Escribano-Ávila, Nina Farwig, Daniel García, Juan C. Illera, Pedro Jordano, Przemysław Kurek, Sascha Rösner, Emilio Virgós & William J. Sutherland

*Correspondence to: juanpe.varo@uca.es

Appendix (pages 2–10)

Appendix S1. Supplementary Material and Methods.

Tables (pages 11–18)

Table S1. Name and characteristics of the study landscapes.

Table S2. Number of samples from DNA barcoding analysis in the forest and matrix of the study landscapes.

Table S3. Incidence matrix of bird and mammal species identified through DNA barcoding as seed dispersers of fleshy-fruited plants in the study landscapes.

Table S4. Incidence matrix of seed species of fleshy-fruited plants sampled in the study landscapes.

Table S5. Data on seed mass and height of plant species collated for this study.

Table S6. Results of GLMMs testing for differences between forest and matrix in Hill-diversity of frugivores, seeds and interactions, and network-level interaction complementarity (H_2').

Table S7. Results of GLMMs testing for differences between forest and matrix in the relative contribution of the main frugivore families.

Table S8. Results of Principal Components Analysis (PCA) accounting for the variability in the relative contribution of the six main families of frugivores to community-wide seed rain in the forest and matrix of the study landscapes.

Figures (pages 19–28)

Figure S1. Photographs illustrating the physiognomy of the forest and matrix habitats in the study landscapes, as well as different aspects of our sampling of avian and mammalian seed dispersal.

Figure S2. Distribution of distances from seed traps placed in the forest (green) and matrix (yellow) of the study landscapes (panels) to the nearest forest edge.

Figure S3. Sample coverage and sampling completeness of frugivore species, seed species and pairwise interactions in the forest and matrix of the study landscapes.

Figure S4. Community-wide seed rain in the forest and matrix of the study landscapes mediated by birds and mammals.

Figure S5. Seed-dispersal networks sampled in the forest and matrix of the study landscapes.

Figure S6. Changes in seed communities in relation to changes in frugivore communities.

Figure S7. Relative contribution to community-wide seed rain in the forest and matrix of the study landscapes of mammals, *Erithacus rubecula* vs. other Muscicapidae species, and Palearctic vs. Afro-Palearctic migrants.

Figure S8. Posterior distribution of estimates for the matrix (habitat) effect on CWM traits of frugivores and plants fitted by generalized joint attribute modeling.

References (pages 29–30)

Appendix S1. Supplementary Materials and Methods

(a) Study design

We conducted our study in forest and matrix habitats of seven fragmented landscapes located in Spain, UK, Germany, Italy and Poland (Fig. 2a), distributed across the Mediterranean and temperate biomes of Europe (Table S1). Thus, our study comprised a paired design as we sampled in paired habitats within landscapes (Fig. 1). The study landscapes ranged from 1 to 3.8 km², were located between 45 and 630 m asl. (see details in Table S1), and comprised one or a few forest patches surrounded by an agricultural matrix of arable land and/or cattle pastures with isolated trees (Fig. 2b). Hereafter, we refer to these two main habitats within each landscape as ‘forest’ and ‘matrix’, respectively. All forest patches and the isolated trees were dominated by broadleaved species, mainly belonging to the genera *Quercus*, *Fraxinus*, *Fagus*, *Betula*, *Acer* and *Salix* (see details in Table S1). Six of these landscapes were studied for one year (June 2016 to June 2017) and one landscape (Garrapilos, southern Spain) for two complete years (October 2013 to October 2015; Table S1). Thus, our sampling covered the entire fruiting periods of all local fleshy-fruited species.

(b) Sampling frugivore-mediated seed dispersal

We sampled community-wide seed dispersal by frugivores in the forest and matrix of the study landscapes using seed traps and fixed transects. Seed traps constitute the standard procedure to sample avian seed-dispersal beneath plant canopies and perching sites (e.g., 1, 2), whereas transects are very useful to sample both avian seed-dispersal in canopy-free areas (e.g., 3) and mammalian seed dispersal (e.g., 4). Through both sampling methods, we can obtain seed-rain densities (number of seeds per unit area) and sample defecated or regurgitated seeds for subsequent DNA barcoding analysis (see next section) to identify the frugivore species responsible for seed dispersal (3, 5). For seed traps, we used 0.22-m²

plastic trays covered with wire mesh to prevent post-dispersal seed predation. We placed 20–24 seed traps in the forest of each landscape ($n = 146$ across landscapes), beneath the canopy of different trees and shrubs not bearing fleshy fruits (see details in Table S1). In the matrix, we placed 20–34 seed traps per landscape ($n = 178$ across landscapes), of which 20–29 were beneath the canopy of isolated trees without fleshy fruits (mainly beneath *Quercus*, *Salix* and *Fraxinus* trees; Fig. 2b and Table S1). In addition, in six of the seven landscapes, we placed 4–5 seed traps under different electricity/telephone pylons (Fig. 2b), which are anthropogenic perches that receive bird-mediated seed dispersal (see 5, 6). We sampled under pylons in all landscapes except in Cabañeros (central Spain), where these infrastructures were absent. In the electricity pylons of Garrapilos, instead of the seed traps described above, we used the concrete-made base (0.6 m^2) of the pylons to sample avian seed dispersal (5). In the forest, distances from seed traps to the nearest forest edge ranged from 1 to 254 m (median = 40 m) and most distances (82%) were under 100 m (see Fig. S2); these distances are very representative in European forests (7). In the matrix, distances from seed traps to the nearest forest edge ranged from 4 to 438 m (median = 110 m) and most distances (95%) were less than 300 m (Fig. S2). Across landscapes, we monitored a total of 324 seed traps ($n = 40$ –58 per landscape). The fact that all seed traps were placed beneath trees and shrubs (or structures) not bearing fleshy fruits implied that all seeds arriving to them were dispersed by birds horizontally away from their source plants. We monitored the seed traps through periodic surveys (fortnightly) in which we sampled bird-dispersed seeds for DNA barcoding analysis and counted the number of seeds of different plant species to calculate seed-rain densities (i.e., seeds per m^2). Additionally, we used the routes adopted for periodically surveying the seed traps as fixed belt transects (1-m wide), whose length ranged between 559 and 4032 m in the forest ($\sim 10,540$ m across landscapes), and between 1610 and 5077 m in the matrix ($\sim 20,340$ m across landscapes). We monitored

a total of 2630–9110-m² of belt transects per landscape fortnightly, where we also searched for frugivore-dispersed seeds and sampled them for subsequent DNA barcoding analysis. We mainly used these transects to sample mammalian seed dispersal (i.e., scats with seeds), which in Europe is mostly mediated by medium-sized carnivores like foxes and martens (8–10). In addition, these transects served us to complement the sampling of avian seed-dispersal in canopy-free areas, where seed rain is extremely low (e.g., 3, 5, 11) and post-dispersal seed predation is typically low due the lack of shelters for rodents (12).

(c) Frugivore and seed identification

We used DNA barcoding analysis of the mitochondrial cytochrome c oxidase subunit I (COI) gene to identify the animal species responsible for the seed-dispersal events, as DNA of animal origin can be extracted from the surface of defecated or regurgitated seeds (3). We sampled individual seeds or droppings with seeds found in seed traps and transects for analysis by pushing them with a minimum of handling into sterile tubes with the aid of the tube cap. We used 1.5- or 2.0-ml tubes for sampling bird droppings and 100-ml tubes for mammal scats. Tubes were labelled and stored in a freezer at –20 °C until DNA extraction. Some frugivore-dispersed seeds visually detected outside the transects were also sampled for DNA barcoding analysis with the aim of increasing sample sizes, particularly for locally rare plant species; in Garrapilos, we also used samples collected from additional seed traps that were used in previous studies (see details in Table S1). Instead, we generally collected a subsample of the droppings when seed traps or transects received many seeds of certain plant species; e.g., ~40% of the hyper-abundant *Pistacia lentiscus* seeds in Garrapilos (13). Note that these sampling strategies did not affect the seed-rain densities recorded in seed traps and transects; they only affected to the samples used for DNA barcoding analysis, which aimed at identifying the relative contribution of frugivore species to seed dispersal (see section ‘Seed-dispersal networks in forest and matrix’).

Detailed laboratory procedures for DNA extraction, PCR, sequencing and species identification of avian seed dispersers can be found in the Supplementary Methods of González-Varo *et al.* (14) (www.nature.com/articles/s41586-021-03665-2#Sec20). For mammalian samples, we followed the protocol and primers described in Alcaide *et al.* (15) to selectively amplify a 758-bp fragment of the vertebrate mitochondrial COI. For mammalian DNA extraction, we rubbed the surface of faeces with a sterile cotton swab soaked in phosphate buffer to sample DNA while minimizing non target vertebrate DNA (e.g., animal preys of carnivorous mammals). For amplification, we used a nested PCR reaction using the primers M13 and BCV-RV2 on the ‘M13BC-FW/BCV-RV1’ amplicon, as this procedure improved successful amplification of suitable positives for sequencing.

Resulting sequences were identified at the species level based on best sequence matches in the ‘BARCODE OF LIFE DATA’ identification system (BOLD; 16) (www.boldsystems.org), typically at a 98–100% similarity (see Supplementary Fig. 1 in 14). We analyzed 3313 samples containing 15,260 seeds of which we successfully identified the frugivore species in 3063 samples (127–1770 per landscape) containing 14,683 seeds (245–9917 per landscape; see Table S2). We obtained an overall identification success of 92.5% (i.e., PCR failure occurred in 7.5% of samples). The 3063 samples included 3093 interaction events between ‘frugivore–seed’ species pairs because some samples (droppings) contained multiple seed species. In three mammal samples (scats) that failed to amplify, we inferred the species according to scat shape and size (9) and to the mammal species successfully identified in the landscape. All barcoding sequences obtained in the present study ($n = 3060$) are publicly available in the data file ‘ML_interactions_dna_barcoding_samples.csv’ deposited at the DRYAD repository (<https://doi.org/10.5061/dryad.1c59zw427>).

After the extraction of animal DNA, we visually identified seed species according to their morphology. To do so, we compared the seeds against a personal reference collection (owned by J.P.G.-V.) and pictures from a guide of seeds of European fleshy-fruited species that includes plants from the Mediterranean and temperate biomes (17). The exceptions were the seed species of 11 samples for which we combined DNA barcoding analysis using chloroplast *MaturaseK* gene (*matK*) (18) to obtain a short list of species from the best sequence matches in BLAST (19), and visual identification of candidate fleshy-fruited plant species that were present around the study landscapes (see details in 14). All seed samples are stored by J.P.G.-V. at the Laboratory of Botany in the University of Cádiz, and plant sequences are publicly available in the data file ‘ML_interactions_dna_barcoding_samples.csv’ deposited at the DRYAD repository (<https://doi.org/10.5061/dryad.1c59zw427>).

(d) Seed-dispersal networks in forest and matrix

We obtained seed-dispersal networks from the forest and matrix of the study landscapes by using the interaction-level seed rain (sr_{ij}) as the weight of pairwise interactions (expressed as seeds per m²) between each plant species i and each frugivore species j (e.g., 14). We used DNA barcoding identifications to calculate the relative contribution (f_{ij}) of each frugivore species j to the seed-rain density of each plant species i in the forest and matrix of the study landscapes. We did so separately for avian and mammalian frugivores using data from seed traps (birds) and transects (mammals), respectively. We calculated these relative contributions as $f_{ij} = n_{\text{DNA-}ij} / n_{\text{DNA-}i}$, where $n_{\text{DNA-}i}$ is the total number of DNA-barcoded seeds of plant species i , and $n_{\text{DNA-}ij}$ the total number of DNA-barcoded seeds of plant species i dispersed by frugivore species j . We then estimated the seed-rain density of plant species i dispersed by frugivore species j in forest and matrix as $sr_{ij} = sr_i \times f_{ij}$, where sr_i is the seed-rain density (seeds per m²) of plant species i measured in seed traps (average

across seed traps) or transects (total seed rain per single fixed transect); for the landscape of Garrapilos, we averaged data across the two study years. In the case of seed traps, we did this process differentiating between natural (canopy) and artificial (pylons) perches in the matrix (i.e., f_{ijp} and sr_{ijp}), for which we calculated seed-rain density as a weighted mean of sr_{ijp} values across p perch types, using the proportion of seed traps beneath tree canopies (0.80–0.85) and pylons (0.15–0.20) as weighting factor (see similar procedures in 13, 14, 20). We then merged data from seed traps (birds) and transects (mammals) so that the result (sr_{ij}) was the seed-rain density of plant species i dispersed by frugivore species j in the forest and matrix of each landscape. We used this interaction-level seed rain (sr_{ij}) as the weight of pairwise interactions (w_{ij} , expressed as seeds per m²) between plant and frugivore species in seed-deposition networks at forest and matrix (e.g., 14). In six interactions where the disperser was a bird, seed rain of the plant i was 0 in the seed traps (i.e., rare species) but both the plant and the interaction were sampled in the transect. In these cases, we used the seed rain by birds of these plant species in transects, which ranged between 1/200th and 1/10th parts of the minimum species-level seed-rain density recorded in seed traps in forest or matrix.

(e) Animal and plant traits

We obtained trait data for the frugivore and plant species identified in the study landscapes. We focused on traits that are expected to act as response traits, determining sensitivity to the open anthropogenic matrix, and/or as effect traits, favoring the realization of seed-dispersal interactions) (21, 22). For frugivores, we focused on three traits: body mass, hand-wing index of birds ($\text{HWI} = 100 \times \text{Kipp's distance/wing length}$), and migratory status. Body mass is typically both a response and an effect trait because it is related to susceptibility to defaunation and capacity to disperse large seeds over long distances (e.g., 23). HWI is a measure of wing pointedness, a proxy for wing aspect ratio and flight efficiency in birds,

and is positively related to flight strength and ability to cross open habitats (24, 25). We obtained species-level mean body mass and mean HWI of birds from the AVONET database (26). For mammals, we obtained species-level mean body mass from EltonTraits 1.0 (27). The migratory status of frugivore species characterizes whether their occurrence in the landscape is permanent or seasonal. However, Palearctic migrants (birds that breed in Europe and winter in southern Europe and Africa north of the Sahara) are often partial migrants, that is, only a fraction of their populations migrates while the other fraction behaves as resident; all Afro-Palearctic migrants (birds that breed in Europe and winter in sub-Saharan Africa) are fully migratory (14, 28). We used published information (mainly taken from bird atlases) characterizing the proportion of migrants (P_{migrants}) in the frugivorous bird species at the study landscapes (P_{migrants} can vary geographically within species) by means of a semiquantitative variable: 0, non-migrant population; 0.1, only a minor fraction migrates; 0.25, a larger fraction migrates but non-migrants prevail; 0.5, roughly half of the population migrates; 0.75, migrants prevail; 0.9, only a minor fraction does not migrate; 1: the whole population migrates (see details in 14) (data available at <https://doi.org/10.5061/dryad.15dv41nx3>). All identified mammals (foxes and martens) were non-migrant species, thereby $P_{\text{migrants}} = 0$.

For plants, we focused on four traits: seed mass, plant height, fruiting dates and type of occurrence in the study landscapes. The first three traits are traits directly involved in interactions with frugivores through morphological trait matching or through spatial and temporal overlap (29). The fourth trait characterizes whether the species occurrence in the landscape is natural or anthropogenic (i.e., exotic and planted plants), which we consider as a proxy of dispersal from cultivated, ornamental and invasive plants of the matrix (6, 30, 31) (Fig. 1c). We obtained the average individual seed mass (mg) at the species level from Torroba Balmori *et al.* (17) for most species (70%), and from other data sources (13, 32,

33) and our own data for the remaining species (see details in Table S5). We obtained average plant height at the species level from the TRY database (34) for most species (60%), and from other data sources and our own data for the remaining species (35–38) (see details in Table S5). For the TRY data, we estimated the average plant height after excluding outliers (observations >3 SD away from the species' mean). The average for each species was calculated first within datasets and then within species (39). Regarding the fruiting phenology, we used the bioclimate-level data on start and end fruiting dates (d_{start} – d_{end}) obtained by González-Varo *et al.* (14) for the plant species and bioclimate of the study landscapes (data available at <https://doi.org/10.5061/dryad.15dv41nx3>). The exceptions (3% of data) were the fruiting dates of three species exclusively dispersed by mammals: *Ceratonia siliqua* (dates from 40), *Prunus cerasifera* (dates imputed from *Prunus domestica*) and *Pyrus* sp. (dates imputed from *Pyrus amygdaliformis*). The start and end fruiting dates (d_{start} – d_{end}) were obtained on a monthly scale (0–12) at 0.5-month accuracy, where exact values represent the transition between months and half values represent the midpoint within months (for example, 1 = end of January–beginning of February; 1.5 = mid-February). Twelve was added to the end date whenever it belonged to the next calendar year (for example, a dispersal period from mid-November to end of March was expressed as $d_{\text{start}} = 10.5$ and $d_{\text{end}} = 15$ (3 + 12)). Average values (d_{start} and d_{end}) for each 'plant species–bioclimate' combination were obtained after averaging across data sources (see details in 14). In this study, we obtained the midpoint fruiting date as $d_{\text{mid}} = (d_{\text{start}} + d_{\text{end}})/2$; d_{mid} was highly correlated with d_{start} and d_{end} (Pearson's $r = 0.926$ and 0.965 , respectively). Finally, we used a Bernoulli-distributed variable to classify the seed species according to the origin of their adult plants in each landscape (1: exotic or planted; 0: wild and native). We used this distinction because some native species only occurred in the

studied landscapes as planted/cultivated plants (e.g., *Ceratonia siliqua*, *Ficus carica*, *Taxus baccata*, *Vitis vinifera*; Table S4).

Table S1. Name and characteristics of the study landscapes: geographical coordinates in decimal degrees, main fleshy-fruited species and tree species in the forest, and main species of isolated trees in the matrix. Six landscapes were sampled all year-round for one year (2016–2017), while Garrapilos was sampled for two years (2013–2015). Seed traps in forest trees and isolated trees were placed beneath the main species listed in the table. In Mediterranean forests, seed traps in shrubs not bearing fleshy fruits were mainly placed beneath the canopy of *Quercus coccifera* and male *Pistacia lentiscus* plants; in Garrapilos, some seed traps were also placed beneath male *Rhamnus alaternus* plants, whereas in Ficuzza seed traps were placed beneath *Quercus pubescens* treelets. In Temperate forests, all seed traps in shrubs not bearing fleshy fruits were mainly placed beneath *Corylus avellana* plants.

Country	Site name	Biome	Altitude (m asl)	Latitude	Longitude	Main fleshy-fruited species	Main species of forest trees	Main species of isolated trees in the matrix
Spain	Garrapilos*	Mediterranean	45–55	36.659	-5.949	<i>Pistacia lentiscus</i> , <i>Olea europaea</i> var. <i>sylvestris</i> and <i>Rhamnus alaternus</i>	<i>Quercus rotundifolia</i> and <i>Quercus suber</i>	<i>Quercus rotundifolia</i> and <i>Quercus suber</i>
Spain	Cabañeros	Mediterranean	620–630	39.321	-4.290	<i>Pistacia terebinthus</i> , <i>Myrtus communis</i> and <i>Phillyrea angustifolia</i>	<i>Quercus rotundifolia</i> , <i>Quercus suber</i> and <i>Fraxinus angustifolia</i>	<i>Quercus rotundifolia</i>
Italy	Ficuzza	Mediterranean	540–600	37.895	13.374	<i>Hedera helix</i> , <i>Crataegus monogyna</i> and <i>Rubus ulmifolius</i>	<i>Quercus pubescens</i>	<i>Fraxinus angustifolia</i>
Spain	Arbazal	Temperate	400–450	43.431	-5.497	<i>Hedera helix</i> , <i>Rhamnus alaternus</i> and <i>Crataegus monogyna</i>	<i>Fraxinus excelsior</i> , <i>Quercus robur</i> and <i>Castanea sativa</i>	<i>Fraxinus excelsior</i> , <i>Betula pubescens</i> and <i>Quercus robur</i>
UK	Bradfield Woods	Temperate	80–95	52.181	0.824	<i>Crataegus monogyna/laevigata</i> , <i>Ilex aquifolium</i> and <i>Hedera helix</i>	<i>Quercus petrae/robur</i> , <i>Betula</i> sp. and <i>Fagus sylvatica</i>	<i>Quercus petrae/robur</i> and <i>Fagus sylvatica</i>
Germany	Bauerbach	Temperate	220–270	50.795	8.823	<i>Sambucus nigra</i> , <i>Crataegus monogyna/laevigata</i> and <i>Cornus sanguinea</i>	<i>Fagus sylvatica</i> , <i>Pinus sylvestris</i> and <i>Quercus petrae/robur</i>	<i>Salix</i> sp. and <i>Populus</i> sp.
Poland	Hebdów	Temperate	185–220	50.143	20.427	<i>Sambucus nigra</i> , <i>Cornus sanguinea</i> and <i>Ligustrum vulgare</i>	<i>Acer platanoides</i> , <i>Fraxinus excelsior</i> and <i>Tilia cordata</i>	<i>Salix alba</i> and <i>Salix × fragilis</i>

* In Garrapilos, we also collected samples for DNA barcoding analysis from additional seed traps used in other studies (3, 5, 13, 20), which did not fit the criteria used here to quantify the seed rain, that is, continuous sampling for the whole study period (i.e., two years in Garrapilos) and be placed beneath plants not bearing fleshy fruits.

Table S2. Number of samples, and number of seeds in those samples, with successful identification of frugivore species after DNA barcoding analysis in the forest and matrix of the study landscapes. Sample sizes are reported separately for avian and mammalian seed dispersal (i.e., bird droppings and mammal scats, respectively). Numbers in brackets denote the total number of samples analyzed and the number of seeds they contained.

Landscape	Avian seed dispersal				Mammalian seed dispersal			
	<i>n</i> samples		<i>n</i> seeds		<i>n</i> samples		<i>n</i> seeds	
	Forest	Matrix	Forest	Matrix	Forest	Matrix	Forest	Matrix
Garrapilos*	1178 (1307)	575 (635)	1452 (1643)	741 (814)	11 (11)	7 (7)	4590 (4590)	3134 (3134)
Cabañeros	72 (76)	51 (51)	72 (76)	72 (72)	2 (2)	2 (2)	17 (17)	84 (84)
Ficuzza	129 (130)	91 (116)	501 (502)	463 (465)	2 (2)	3 (5)	30 (30)	472 (472)
Arbazal	163 (178)	96 (109)	269 (285)	123 (137)	10 (10)	2 (3)	558 (558)	10 (14)
Bradfield Woods	129 (134)	91 (92)	261 (299)	165 (166)	2 (2)	3 (3)	609 (609)	20 (20)
Bauerbach	44 (45)	153 (157)	67 (68)	248 (256)	3 (3)	11 (13)	26 (26)	129 (141)
Hebdów	66 (71)	129 (133)	149 (156)	229 (242)	9 (11)	5 (5)	140 (162)	52 (52)
Total	1781 (1941)	1210 (1293)	2771 (3029)	2041 (2152)	39 (41)	33 (38)	5970 (5992)	3901 (3917)

* The larger samples sizes from Garrapilos reflect that this was the only landscape sampled for two years and that DNA barcoding analysis was also conducted on samples from additional seed traps or directed searches used in other studies (3, 5, 13, 20). The large number of mammal-dispersed seeds in Garrapilos is accounted for several fox scats that contained hundreds of seeds (up to 875 per scat) of *Rubus ulmifolius*.

Table S3. Incidence matrix of bird and mammal species identified through DNA barcoding as seed dispersers of fleshy-fruited plants in the study landscapes (sites) with totals per species and site. Bird species belong to the Order Passeriformes except those from families Columbidae (Columbiformes), Falconidae (Falconiformes), Phasianidae (Galliformes) and Picidae (Piciformes). The mammal species belong to the O. Carnivora. We followed taxonomy from ‘Birds of the World’ (www.birdsoftheworld.org) for birds and the ‘Integrated Taxonomic Information System’ for mammals (www.itis.gov). On average, we identified 15.7 disperser species per landscape and each species was sampled in 2.6 landscapes. Common and widespread species (e.g., *Erithacus rubecula* and *Turdus merula*) were detected in the seven landscapes, while rarer species (e.g., *Turdus torquatus*), species with narrower distribution (e.g., *Cyanopica cooki*) and/or those that only occasional behave as seed dispersers (e.g., *Lanius excubitor*) were identified in fewer landscapes.

Disperser species (family)	Mediterranean			Temperate				Total (n sites per species)
	Garrapilos (Spain)	Cabañeros (Spain)	Ficuzza (Italy)	Arbazal (Spain)	B. Woods (UK)	Bauerbach (Germany)	Hebđów (Poland)	
BIRDS								
1. <i>Alectoris rufa</i> (Phasianidae)	1	0	0	0	0	0	0	1
2. <i>Columba palumbus</i> (Columbidae)	1	0	1	1	1	1	1	6
3. <i>Corvus cornix</i> (Corvidae)	0	0	1	0	0	0	0	1
4. <i>Corvus corone</i> (Corvidae)	0	0	0	1	0	1	0	2
5. <i>Corvus monedula</i> (Corvidae)	1	0	0	0	0	0	0	1
6. <i>Curruca cantillans</i> (Sylviidae)	1	1	0	0	0	0	0	2
7. <i>Curruca communis</i> (Sylviidae)	1	1	0	0	1	1	1	5
8. <i>Curruca hortensis</i> (Sylviidae)	1	1	0	0	0	0	0	2
9. <i>Curruca melanocephala</i> (Sylviidae)	1	1	0	0	0	0	0	2
10. <i>Curruca undata</i> (Sylviidae)	0	1	0	0	0	0	0	1
11. <i>Cyanistes caeruleus</i> (Paridae)	0	1	1	0	0	0	1	3
12. <i>Cyanopica cooki</i> (Corvidae)	0	1	0	0	0	0	0	1
13. <i>Emberiza calandra</i> (Emberizidae)	0	1	0	0	0	0	0	1
14. <i>Erithacus rubecula</i> (Muscicapidae)	1	1	1	1	1	1	1	7
15. <i>Falco tinnunculus</i> (Falconidae)	0	0	0	1	0	0	0	1
16. <i>Ficedula hypoleuca</i> (Muscicapidae)	1	1	0	0	0	0	0	2
17. <i>Fringilla coelebs</i> (Fringillidae)	0	0	1	0	0	0	1	2
18. <i>Garrulus glandarius</i> (Corvidae)	0	0	1	1	0	0	1	3
19. <i>Lanius excubitor</i> (Laniidae)	0	0	0	0	0	0	1	1
20. <i>Luscinia megarhynchos</i> (Muscicapidae)	1	0	0	0	0	0	0	1
21. <i>Muscicapa striata</i> (Muscicapidae)	1	0	0	0	0	0	0	1
22. <i>Oriolus oriolus</i> (Oriolidae)	0	0	0	0	0	0	1	1
23. <i>Parus major</i> (Paridae)	1	0	0	0	0	0	1	2
24. <i>Phasianus colchicus</i> (Phasianidae)	0	0	0	0	1	0	0	1
25. <i>Phoenicurus ochruros</i> (Muscicapidae)	1	0	0	1	0	0	0	2
26. <i>Phoenicurus phoenicurus</i> (Muscicapidae)	1	1	0	0	0	0	0	2
27. <i>Pica pica</i> (Corvidae)	0	0	0	0	0	0	1	1
28. <i>Picus sharpei</i> (Picidae)	1	0	0	1	0	0	0	2
29. <i>Saxicola rubicola</i> (Muscicapidae)	1	0	0	0	0	0	0	1
30. <i>Streptopelia decaocto</i> (Columbidae)	0	0	0	0	0	0	1	1
31. <i>Sturnus unicolor</i> (Sturnidae)	1	1	0	0	0	0	0	2
32. <i>Sturnus vulgaris</i> (Sturnidae)	0	0	1	1	1	1	1	5
33. <i>Sylvia atricapilla</i> (Sylviidae)	1	1	1	1	1	1	1	7
34. <i>Sylvia borin</i> (Sylviidae)	1	0	0	1	0	0	0	2
35. <i>Turdus iliacus</i> (Turdidae)	0	0	0	1	1	0	0	2
36. <i>Turdus merula</i> (Turdidae)	1	1	1	1	1	1	1	7
37. <i>Turdus philomelos</i> (Turdidae)	1	0	1	1	1	1	1	6
38. <i>Turdus pilaris</i> (Turdidae)	0	0	0	0	1	1	1	3
39. <i>Turdus torquatus</i> (Turdidae)	0	0	1	0	0	0	0	1
40. <i>Turdus viscivorus</i> (Turdidae)	0	0	1	1	1	0	0	3
MAMMALS								
41. <i>Martes foina</i> (Mustelidae)	0	1	0	1	0	1	1	4
42. <i>Martes martes</i> (Mustelidae)	0	0	1	1	0	0	1	3
43. <i>Vulpes vulpes</i> (Canidae)	1	1	1	1	1	1	0	6
Total (n disperser species per landscape)	22	16	14	17	12	11	18	–

Table S4. Incidence matrix of seed species of fleshy-fruited plants sampled in the study landscapes with totals per species and landscape. We followed taxonomy from ‘World Flora Online’ (www.worldfloraonline.org). On average, we sampled 14.1 seed species per landscape and each species was sampled in 2.1 landscapes. Underscored numbers indicate anthropogenic occurrence in the landscape as exotic or planted species.

Seed species (family)	Mediterranean biome			Temperate biome				Total (<i>n</i> landscapes per species)
	Garrapilos (Spain)	Cabañeros (Spain)	Ficuzza (Italy)	Arbazal (Spain)	B. Woods (UK)	Bauerbach (Germany)	Hebdów (Poland)	
1. <i>Arum italicum</i> (Araceae)	1	0	0	0	0	0	0	1
2. <i>Arum maculatum</i> (Araceae)	0	0	0	0	1	0	0	1
3. <i>Asparagus acutifolius</i> (Asparagaceae)	1	0	1	0	0	0	0	2
4. <i>Asparagus aphyllus</i> (Asparagaceae)	0	1	0	0	0	0	0	1
5. <i>Bryonia cretica</i> (Cucurbitaceae)	1	1	0	0	0	0	0	2
6. <i>Ceratonia siliqua</i> (Fabaceae)	<u>1</u>	0	0	0	0	0	0	1
7. <i>Cornus sanguinea</i> (Cornaceae)	0	0	0	1	1	1	1	4
8. <i>Crataegus monogyna</i> * (Rosaceae)	1	1	1	1	1	1	0	6
9. <i>Daphne laureola</i> (Thymeleaceae)	0	0	0	0	1	0	0	1
10. <i>Dioscorea communis</i> (Dioscoreaceae)	1	1	1	1	1	0	0	5
11. <i>Euonymus europaeus</i> (Celastraceae)	0	0	0	0	1	1	1	3
12. <i>Ficus carica</i> (Moraceae)	<u>1</u>	0	<u>1</u>	<u>1</u>	0	0	0	3
13. <i>Hedera helix</i> (Araliaceae)	0	0	1	1	1	0	0	3
14. <i>Ilex aquifolium</i> (Aquifoliaceae)	0	0	0	1	1	0	0	2
15. <i>Jasminum fruticans</i> (Oleaceae)	0	1	0	0	0	0	0	1
16. <i>Lonicera etrusca</i> (Caprifoliaceae)	0	1	0	0	0	0	0	1
17. <i>Lonicera periclymenum</i> (Caprifoliaceae)	0	0	0	0	1	0	0	1
18. <i>Morus alba</i> (Moraceae)	0	0	<u>1</u>	0	0	0	0	1
19. <i>Morus nigra</i> (Moraceae)	<u>1</u>	0	0	0	0	0	0	1
20. <i>Myrtus communis</i> (Myrtaceae)	1	1	0	0	0	0	0	2
21. <i>Olea europaea</i> – cultivated (Oleaceae)	0	0	<u>1</u>	0	0	0	0	1
22. <i>Olea europaea</i> – wild (Oleaceae)	1	0	0	0	0	0	0	1
23. <i>Osyris alba</i> (Santalaceae)	0	1	0	0	0	0	0	1
24. <i>Phillyrea angustifolia</i> (Oleaceae)	0	1	0	0	0	0	0	1
25. <i>Pistacia lentiscus</i> (Anacardiaceae)	1	1	0	0	0	0	0	2
26. <i>Pistacia terebinthus</i> (Anacardiaceae)	0	1	0	0	0	0	0	1
27. <i>Prunus avium</i> (Rosaceae)	0	0	0	1	0	1	1	3
28. <i>Prunus cerasifera</i> (Rosaceae)	0	0	0	0	0	0	<u>1</u>	1
29. <i>Prunus domestica</i> (Rosaceae)	0	0	0	<u>1</u>	<u>1</u>	<u>1</u>	<u>1</u>	4
30. <i>Prunus spinosa</i> (Rosaceae)	0	0	0	1	1	1	0	3
31. <i>Pyrus amygdaliformis</i> (Rosaceae)	0	0	1	0	0	0	0	1
32. <i>Pyrus</i> sp. (Rosaceae)	0	0	0	0	0	0	<u>1</u>	1
33. <i>Rhamnus alaternus</i> (Rhamnaceae)	1	0	0	1	0	0	0	2
34. <i>Rhamnus lycioides</i> (Rhamnaceae)	1	1	0	0	0	0	0	2
35. <i>Ribes rubrum</i> (Grossulariaceae)	0	0	0	0	0	0	1	1
36. <i>Rosa canina</i> /sp.** (Rosaceae)	0	1	1	0	1	1	0	4
37. <i>Rubia peregrina</i> (Rubiaceae)	0	1	1	1	0	0	0	3
38. <i>Rubus plicatus/ulmifolius</i> *** (Rosaceae)	1	0	1	1	1	1	1	6
39. <i>Ruscus aculeatus</i> (Asparagaceae)	0	0	1	1	0	0	0	2
40. <i>Sambucus nigra</i> (Adoxaceae)	0	0	0	1	1	1	1	4
41. <i>Smilax aspera</i> (Smilacaceae)	1	0	0	1	0	0	0	2
42. <i>Solanum dulcamara</i> (Solanaceae)	0	0	0	0	<u>1</u>	0	<u>1</u>	2
43. <i>Solanum nigrum</i> (Solanaceae)	0	1	0	0	0	0	0	1
44. <i>Sorbus aucuparia</i> (Rosaceae)	0	0	0	0	0	1	1	2
45. <i>Symphoricarpos albus</i> (Caprifoliaceae)	0	0	0	0	0	0	<u>1</u>	1
46. <i>Taxus baccata</i> (Taxaceae)	0	0	0	0	<u>1</u>	0	0	1
47. <i>Viburnum opulus</i> (Adoxaceae)	0	0	0	0	0	1	0	1
48. <i>Vitis vinifera</i> (Vitaceae)	0	<u>1</u>	<u>1</u>	0	0	0	<u>1</u>	3
Total (<i>n</i> seed species per landscape)	15	16	13	15	16	11	13	–

* Species aggregate of *Crataegus monogyna/laevigata* in Bradfield Woods and Bauerbach.

** Species aggregate that includes *Rubus plicatus/caesius* in Bradfield Woods and & Bauerbach.

*** Species aggregate that includes several *Rosa* species along with *R. canina*.

Table S5. Data on mean seed mass and mean height of plant species collated for this study. Most data on seed mass is from a guide of seeds of fleshy fruits (17) complemented by other references and data from the authors and colleagues. Most data on plant height is from the TRY database (34) complemented by other references and data from the authors. Our plant height data was obtained by JPGV along transects characterizing the vegetation of Mediterranean forest patches of SW Iberia (41); height data of *Pyrus amygdaliformis* obtained by RSB in Ficuzza. In few cases, seed mass (*Pyrus* sp.) and plant height (*Arum italicum*, cultivated *Olea europaea* and *Pyrus* sp.) were imputed from sister taxa. Plant height data from two sources (e.g., *Pistacia terebinthus*) results from the mean value across both sources.

Seed species (family)	Seed mass		Plant height	
	Mean (mg)	Source	Mean (m)	Source
1. <i>Arum italicum</i>	38.6	(17)	0.36	Imputed from <i>Arum maculatum</i>
2. <i>Arum maculatum</i>	45.0	This study	0.36	(34)
3. <i>Asparagus acutifolius</i>	40.0	(33)	0.55	This study
4. <i>Asparagus aphyllus</i>	39.0	(33)	0.58	This study
5. <i>Bryonia cretica</i>	13.7	(17)	2.06	This study
6. <i>Ceratonia siliqua</i>	180.0	(32)	7.00	(35)
7. <i>Cornus sanguinea</i>	56.6	(17)	4.19	(34)
8. <i>Crataegus monogyna</i>	78.7	(17)	7.35	(34)
9. <i>Daphne laureola</i>	14.3	(17)	0.93	(34)
10. <i>Dioscorea communis</i>	21.2	(17)	1.96	(This study, 34)
11. <i>Euonymus europaeus</i>	29.2	(17)	3.91	(34)
12. <i>Ficus carica</i>	0.5	(17)	6.50	(34)
13. <i>Hedera helix</i>	28.9	(17)	11.15	(34)
14. <i>Ilex aquifolium</i>	25.6	(17)	11.47	(34)
15. <i>Jasminum fruticans</i>	29.7	(17)	2.05	(34)
16. <i>Lonicera etrusca</i>	8.7	(17)	2.00	(35)
17. <i>Lonicera periclymenum</i>	5.5	(17)	4.70	(34)
18. <i>Morus alba</i>	1.8	(17)	13.33	(34)
19. <i>Morus nigra</i>	1.0	This study	11.63	(34)
20. <i>Myrtus communis</i>	10.8	(42)	1.51	This study
21. <i>Olea europaea</i> – cultivated	432.0	This study	6.60	Imputed from wild <i>Olea europaea</i>
22. <i>Olea europaea</i> – wild	221.0	(33)	6.60	(5)
23. <i>Osyris alba</i>	90.7	(17)	0.69	This study
24. <i>Phillyrea angustifolia</i>	18.4	(17)	1.98	This study
25. <i>Pistacia lentiscus</i>	16.9	(13)	1.94	This study
26. <i>Pistacia terebinthus</i>	24.4	(17)	5.05	(37, 38)
27. <i>Prunus avium</i>	197.6	(17)	8.33	(36)
28. <i>Prunus cerasifera</i>	496.0	This study	7.75	(34)
29. <i>Prunus domestica</i>	436.0	This study	7.93	(34)
30. <i>Prunus spinosa</i>	183.9	(17)	3.07	(34)
31. <i>Pyrus amygdaliformis</i>	57.0	This study	4.05	This study
32. <i>Pyrus</i> sp.	57.0	Imputed from <i>Pyrus amygdaliformis</i>	4.05	Imputed from <i>Pyrus amygdaliformis</i>
33. <i>Rhamnus alaternus</i>	11.4	(17)	2.71	This study
34. <i>Rhamnus lycioides</i>	9.9	This study	1.60	This study
35. <i>Ribes rubrum</i>	5.7	(17)	1.62	(34)
36. <i>Rosa canina</i> /sp.	16.1	(17)	2.30	(34)
37. <i>Rubia peregrina</i>	20.2	(17)	0.70	(34)
38. <i>Rubus plicatus/ulmifolius</i>	2.5	(17)	1.98	(34)
39. <i>Ruscus aculeatus</i>	207.6	(17)	0.95	This study
40. <i>Sambucus nigra</i>	2.5	(17)	7.88	(34)
41. <i>Smilax aspera</i> (Smilacaceae)	36.8	(17)	1.98	This study
42. <i>Solanum dulcamara</i>	1.5	(17)	2.07	(34)
43. <i>Solanum nigrum</i>	0.9	This study	0.76	(34)
44. <i>Sorbus aucuparia</i>	4.5	(17)	8.81	(34)
45. <i>Symphoricarpos albus</i>	9.2	(17)	1.92	(34)
46. <i>Taxus baccata</i>	70.1	(17)	19.75	(34)
47. <i>Viburnum opulus</i>	37.0	(17)	3.68	(34)
48. <i>Vitis vinifera</i> *	27.2	(17)	2.00	This study

* Cultivated *Vitis vinifera* plants in vineyards.

Table S6. Generalized linear mixed models (GLMMs) testing for differences between forest and matrix in Hill diversity metrics (species richness and Hill Shannon) describing three components of seed-dispersal networks: (i) frugivore species contributing to seed rain; (ii) plant species dispersed, and (iii) pairwise interactions between frugivore and plant species. (iv) GLMM testing for differences between forest and matrix in network-level interaction complementarity (H_2'). Family distributions are shown (link functions: identity for normal, log for Poisson and logit for beta distributions) along with parameter estimates and the variance of the random factor (i.e., landscape identity; $n = 14$ observations, 7 landscapes \times 2 habitat types per landscape). Significant ($P < 0.05$) model estimates for the ‘Habitat (matrix)’ effect are shown in bold.

GLMMs by response variable (Hill diversity metrics and network-level complementarity)	Family distribution	Conditional model (estimates \pm se)		Random factor (landscape)
		Intercept	Habitat (matrix)	Variance
<i>(i)</i> Frugivore species				
Richness	$y \sim \text{Poisson}(\lambda)$	2.427 \pm 0.124	0.129 \pm 0.153	0.018
Hill-Shannon	$y \sim \text{Gamma}(\mu, \sigma^2)$	1.546 \pm 0.109	0.237 \pm 0.096	0.051
<i>(ii)</i> Plant species (seeds)				
Richness	$y \sim \text{Poisson}(\lambda)$	2.461 \pm 0.110	-0.173 \pm 0.163	6.6×10^{-11}
Hill-Shannon	$y \sim \text{Gamma}(\mu, \sigma^2)$	1.545 \pm 0.141	0.068 \pm 0.126	0.083
<i>(iii)</i> Pairwise interactions				
Richness	$y \sim \text{Poisson}(\lambda)$	3.405 \pm 0.094	-0.135 \pm 0.100	0.029
Hill-Shannon	$y \sim \text{Gamma}(\mu, \sigma^2)$	2.514 \pm 0.136	-0.020 \pm 0.122	0.077
<i>(iv)</i> Network specialization				
H_2' : interaction complementary	$y \sim \text{Beta}(\mu, \phi)$	0.017 \pm 0.154	0.616 \pm 0.218	0.007

Table S7. Generalized linear mixed models (GLMMs) testing for differences between forest and matrix in the relative contribution to community-wide seed rain by the six main families of seed dispersers (i.e., those with a mean relative contribution across landscapes > 5% in forest or matrix). Proportions were modelled using a beta distribution and logit link. Parameter estimates and the variance of the random factor (i.e., landscape identity; $n = 14$ observations, 7 landscapes \times 2 habitat types per landscape) are shown. Significant model estimates ($P < 0.05$) for the effect of ‘Habitat’ (matrix) are shown in bold.

GLMMs by response variable (i.e., relative contributions to community wide seed rain by different frugivore families)	Family distribution	Conditional model (estimates \pm se)		Random factor (landscape)
		Intercept	Habitat (matrix)	Variance
Columbidae*	$y \sim \text{Beta}(\mu, \phi)$	-3.037 ± 0.092	0.749 ± 0.282	0.010
Corvidae	$y \sim \text{Beta}(\mu, \phi)$	-2.474 ± 0.323	0.326 ± 0.317	0.234
Muscicapidae	$y \sim \text{Beta}(\mu, \phi)$	-1.345 ± 0.294	-1.043 ± 0.261	0.435
Sturnidae**	$y \sim \text{Beta}(\mu, \phi)$	-3.223 ± 0.040	1.792 ± 0.347	7.3×10^{-7}
Sylviidae	$y \sim \text{Beta}(\mu, \phi)$	-1.617 ± 0.430	-0.553 ± 0.221	1.128
Turdidae	$y \sim \text{Beta}(\mu, \phi)$	0.033 ± 0.386	-0.161 ± 0.280	0.785

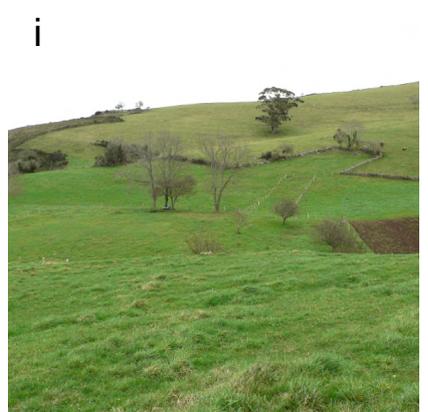
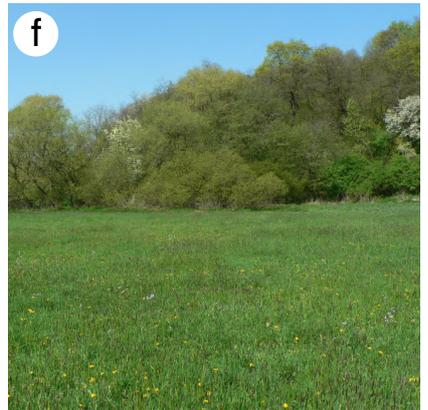
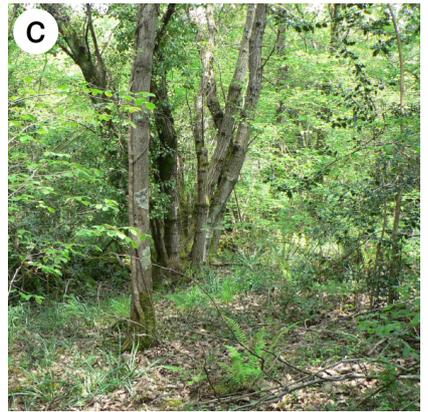
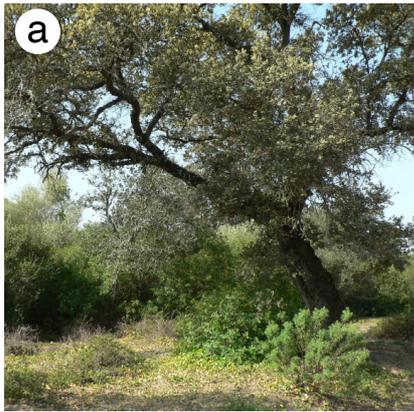
* Dispersion parameter ϕ of the beta distribution was allowed to vary with habitat type (intercept = 6.274 ± 1.632 ; Habitat [matrix] = -3.175 ± 1.646) because the model had a better fit ($\Delta\text{AIC} = -10$) relative to a model with fixed ϕ .

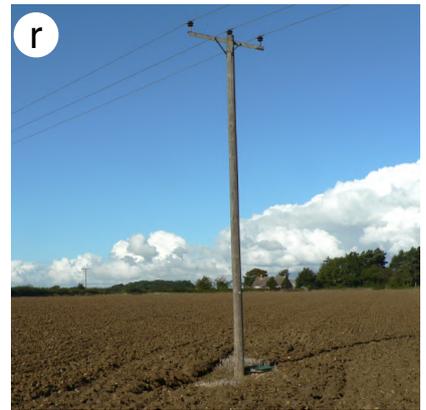
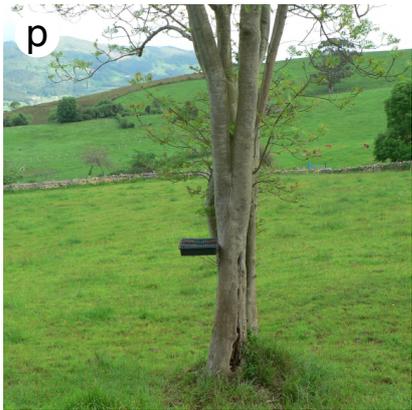
** Dispersion parameter ϕ of the beta distribution was allowed to vary with habitat type (intercept = 7.813 ± 0.528 ; Habitat [matrix] = -5.924 ± 0.746) because the model had a better fit ($\Delta\text{AIC} = -27$) relative to a model with fixed ϕ .

Table S8. Results of Principal Components Analysis (PCA) accounting for the variability in the relative contribution of the six main families of frugivores to community-wide seed rain in the forest and matrix of the study landscapes. PCA factor scores were obtained from the three first rotated eigenvectors, which explained a cumulative variance explained of 83.5%. The percentage of variance accounted for by each eigenvector, as well as the matrix of variable loadings are shown. We tested for differences between forest and matrix in the PCs by means of linear mixed models (LMMs) that included ‘landscape identity’ as random factor (random intercepts). PC1 was significantly higher in the matrix ($\chi^2 = 17.72$, $P < 2 \times 10^{-16}$), whereas PC2 ($\chi^2 = 3.18$, $P = 0.075$) and PC3 ($\chi^2 = 4.41$, $P = 0.036$) were respectively, marginally significantly and significantly higher in the forest.

Main families	Principal Components (% variance explained)		
	PC1 (40.8%)	PC2 (27.3%)	PC3 (15.4%)
Columbidae	0.261	-0.525	0.352
Corvidae	0.202	0.547	-0.528
Muscicapidae	-0.522	0.079	0.313
Sturnidae	0.214	-0.557	-0.548
Sylviidae	-0.566	-0.041	-0.146
Turdidae	0.503	0.326	0.423

Figure S1. Photographs illustrating the physiognomy of the forest and matrix habitats in the study landscapes, as well as different aspects of our sampling of avian and mammalian seed dispersal. (a-c) Interior of a Mediterranean (a) and two temperate (b, c) forests. (d-f) Forest edges adjoining open agricultural matrices dominated by arable crops and/or cattle pastures. (g-l) Isolated trees (non-fleshy-fruited species) in the matrix of the study landscapes used to place seed traps and sample avian seed dispersal. (m-o) Seed traps beneath tree (m) and shrub (n, o) canopies to sample avian seed dispersal in the forest of the study landscapes. (p-r) Seed traps placed beneath the canopy of isolated trees (p, q; i.e., natural perches) and under a pylon (r; i.e., anthropogenic perches) to sample avian seed dispersal in the matrix of the study landscapes. (s-u) Examples of bird-dispersed seeds found in seed traps (s: a defecated *Crataegus monogyna* seed; t: many defecated *Rubus plicatus/ulmifolius* seeds; u: a regurgitated *Ilex aquifolium* seed). (v) Monitoring a seed trap at an isolated tree in the matrix. (w-x) Mammal scats detected in transects containing seeds of *Rubus plicatus/ulmifolius* (w) and (x) *Prunus avium*.





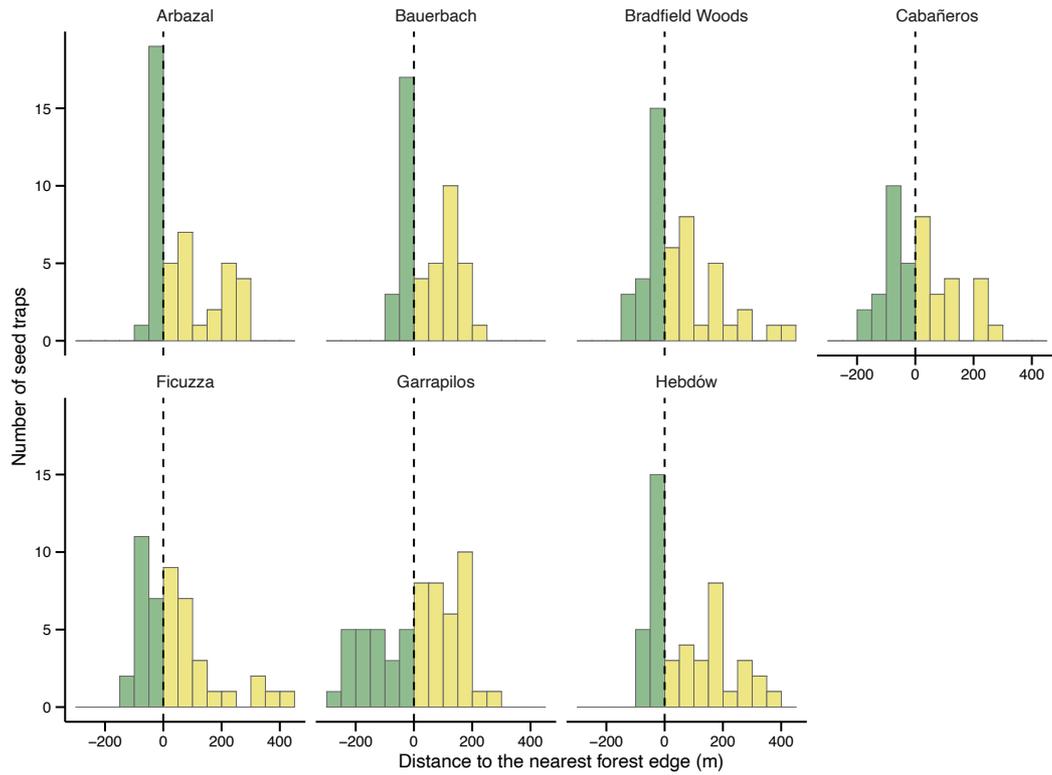


Figure S2. Distribution of distances from seed traps placed in the forest (green) and matrix (yellow) of the study landscapes (panels) to the nearest forest edge (bin width = 50 m). Distances were obtained in QGIS v.3.26.1 (43) using satellite images and the seed-traps coordinates. In the forest, distances from seed traps to the nearest forest edge ranged from 1 to 254 m (median = 40 m) and most (82%) were under 100 m; these distances are very representative of European forests (7). In the matrix, distances from seed traps to the nearest forest edge ranged from 4 to 438 m (median = 110 m) and most (95%) were less than 300 m.

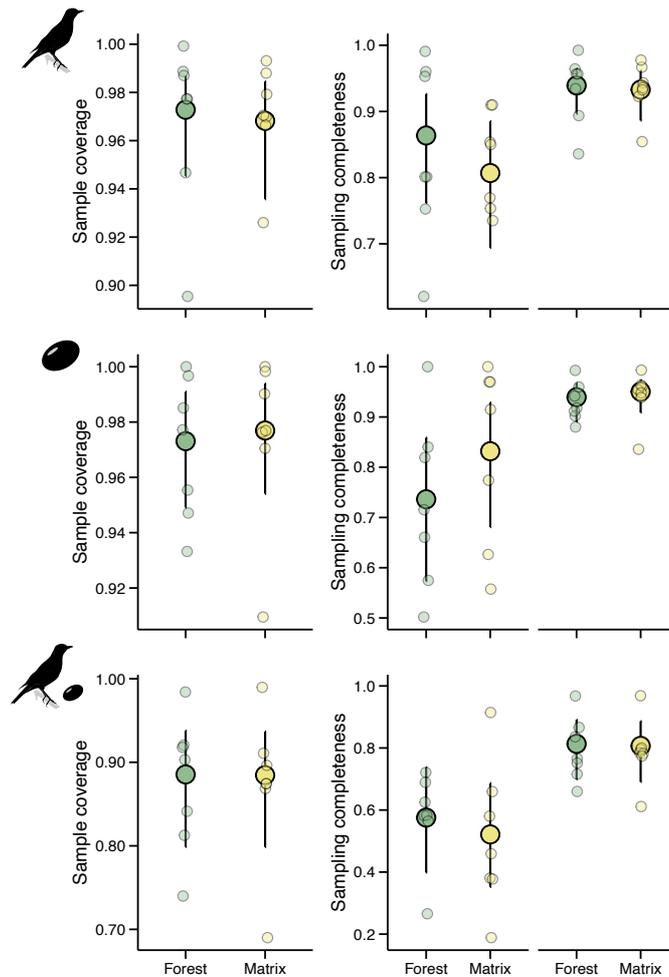


Figure S3. Sample coverage and sampling completeness of species of frugivores (upper panels) and seeds (middle panels), as well as of unique pairwise interactions (lower panels), in the forest and matrix of the study landscapes. In our case, sample coverage is the proportion of seed-dispersal events (i.e., droppings/scats with seeds) including species or interactions present in our DNA-barcoding samples (44), whereas sampling completeness is the percent asymptotic richness and Hill-Shannon detected by our samples (i.e., $100 \times \text{observed diversity} / \text{asymptotic diversity estimate}$) (45). Large circles and bars denote back-transformed means \pm 95% confidence intervals estimated by GLMMs with beta distribution and logit link ('landscape' included as random factor), whereas small circles denote observed values. Sample coverage values were high and almost identical in forest and matrix, with non-significant differences between habitats for seed dispersers ($\sim 97\%$), seeds ($\sim 97.5\%$) and pairwise interactions ($\sim 88.5\%$) (all Wald $\chi^2 = 0.004\text{--}0.180$ and all $P = 0.671\text{--}0.951$). Sampling completeness of Hill-Shannon diversity was higher than sampling completeness of richness. Importantly, sampling completeness values were also almost identical in forest and matrix, with non-significant differences between habitats for seed dispersers, seeds and pairwise interactions for the two Hill diversity metrics (all Wald $\chi^2 = 0.075\text{--}1.109$ and all $P = 0.292\text{--}0.785$). For these analyses, we built three incidence matrices with the DNA-barcoding samples with successful disperser identification ($n = 3063$ samples including 3093 interactions; some droppings contained multiple seeds species) in the forest and matrix of each landscape: (i) frugivore species, (ii) seed species and (iii) pairwise interactions. In the three matrices, columns were individual samples, whereas rows were frugivore species, seed species and unique pairwise interactions, respectively. We then computed rarefaction and extrapolation analysis proposed by Chao *et al.* (46) for Hill diversity orders $q = 0$ (species richness) and $q = 1$ (Hill-Shannon) using the R package iNEXT package (47).

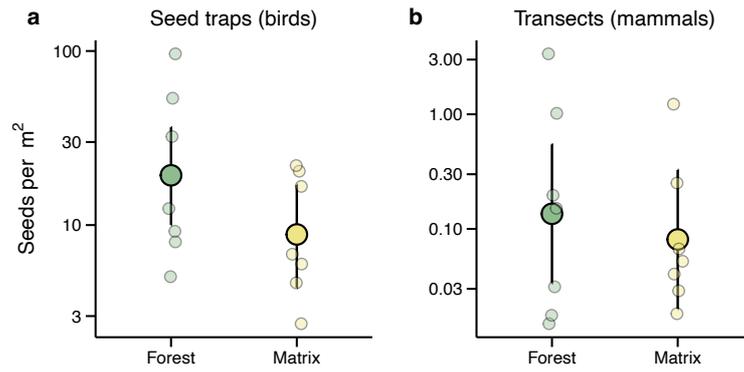


Figure S4. Community-wide seed rain in the forest and matrix of the study landscapes mediated by (a) birds and (b) mammals. Large circles and bars denote back-transformed means \pm 95% confidence intervals estimated by GLMMs, whereas small circles denote observed values. Bird-mediated seed rain sampled in seed traps was significantly higher (more than two times) in forest than in matrix (mean = 19.3 and 8.8 seeds m^{-2} , respectively; Wald $\chi^2 = 21.70$, $P = < 2 \times 10^{-16}$). Mammal-mediated seed rain sampled in transects did not differ significantly between forest and matrix (mean = 13.5 and 8.1 seeds per 100- m^2 , respectively; Wald $\chi^2 = 0.46$, $P = 0.500$). Both models were GLMMs with normal distribution and identity link ('landscape' included as random factor) using log-transformed data (seed traps: $\log_{10}[\text{seed rain} + 1]$; transects: $\log[\text{seed rain}]$); note the logarithmic scale in the y axis.

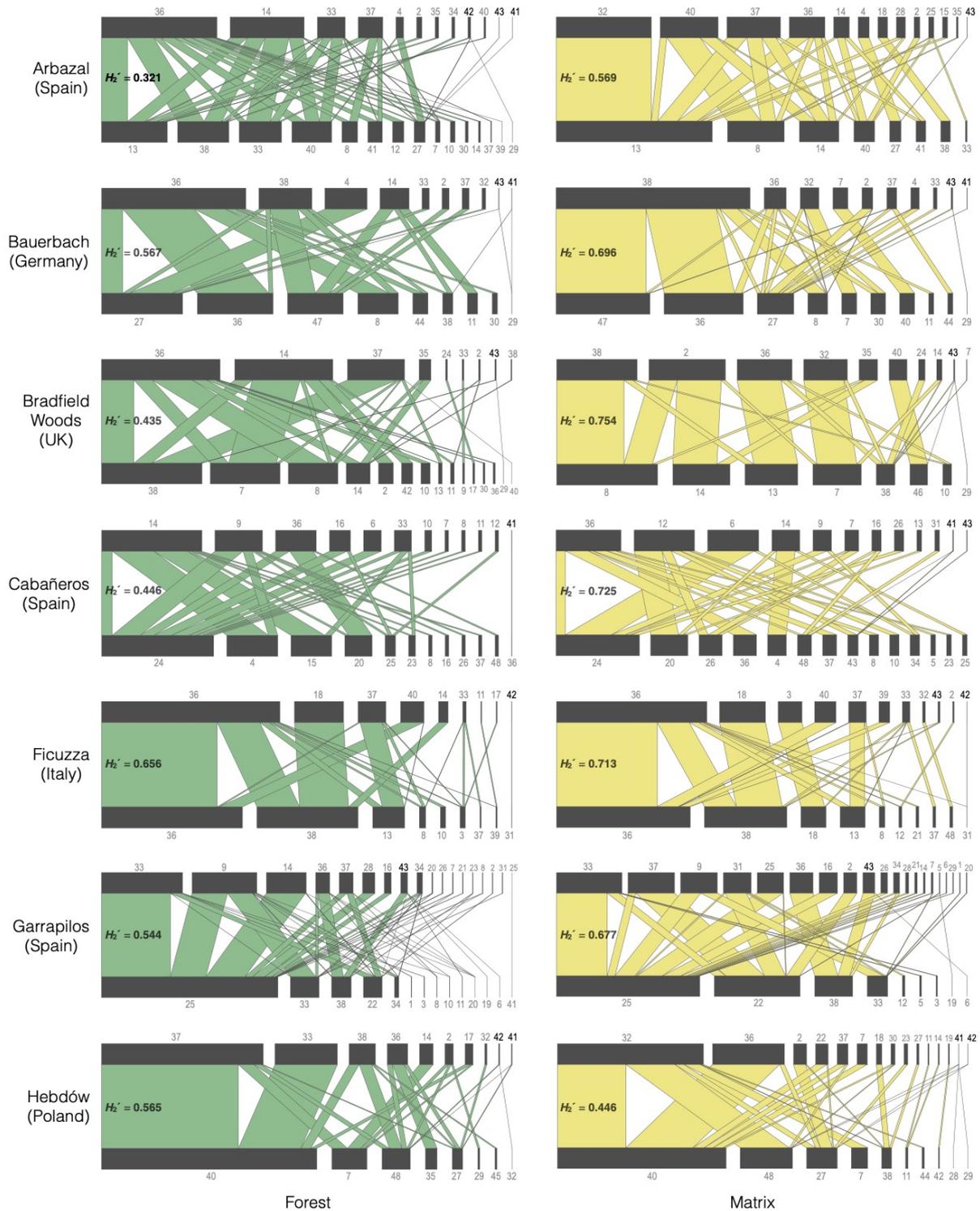


Figure S5. Seed-dispersal networks sampled in the forest and matrix of the study landscapes (rows of panels), with frugivore species represented as the upper nodes (rectangles) and plant species as the lower nodes. Horizontal width of nodes and links is proportional to the seed-rain density contributed by species and pairwise interactions, respectively. Numeric codes in the nodes correspond to the number associated to the frugivore and plant species listed in Tables S3 and S4, respectively. For example, the leftmost link in the ‘Arbazal-forest’ network is the interaction between *Turdus merula* (frugivore species 36 in Table S3) and *Hedera helix* (plant species 13 in Table S4). For frugivores, grey numbers are bird species, while black numbers are mammal species.

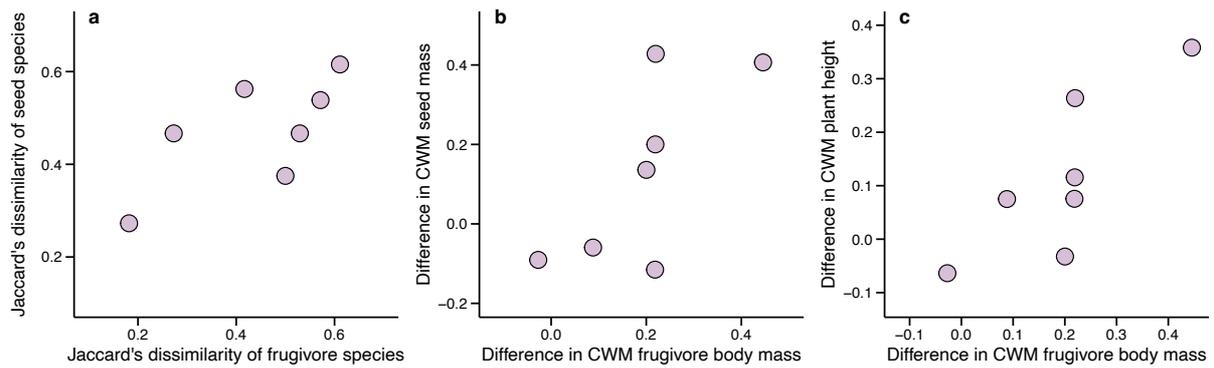


Figure S6. (a) Beta diversity (Jaccard's dissimilarity index) of dispersed seeds between forest and matrix in the seven study landscapes in relation to beta diversity of frugivores (one-sided Pearson test: $r = 0.687$, one-sided $P = 0.044$). (b) Difference between matrix and forest in community-weighted mean (CWM) seed mass of the dispersed seed communities (i.e., $CWM_{matrix} - CWM_{forest}$) in relation to the difference in CWM frugivore body mass (one-sided Pearson test: $r = 0.714$, one-sided $P = 0.036$). (c) Difference between matrix and forest in CWM plant height of the dispersed seed communities in relation to the difference in CWM frugivore body mass (one-sided Pearson test: $r = 0.813$, one-sided $P = 0.013$). CWM traits are \log_{10} -transformed data. We used one-sided tests as we hypothesized these relationships to be positive, that is, (a) the more different the frugivore assemblages, the more different the composition of the seed communities they disperse; and (b-c) the more different the average frugivore size between forest and matrix, the more different the average seed mass and plant size (height) of the dispersed plant species.

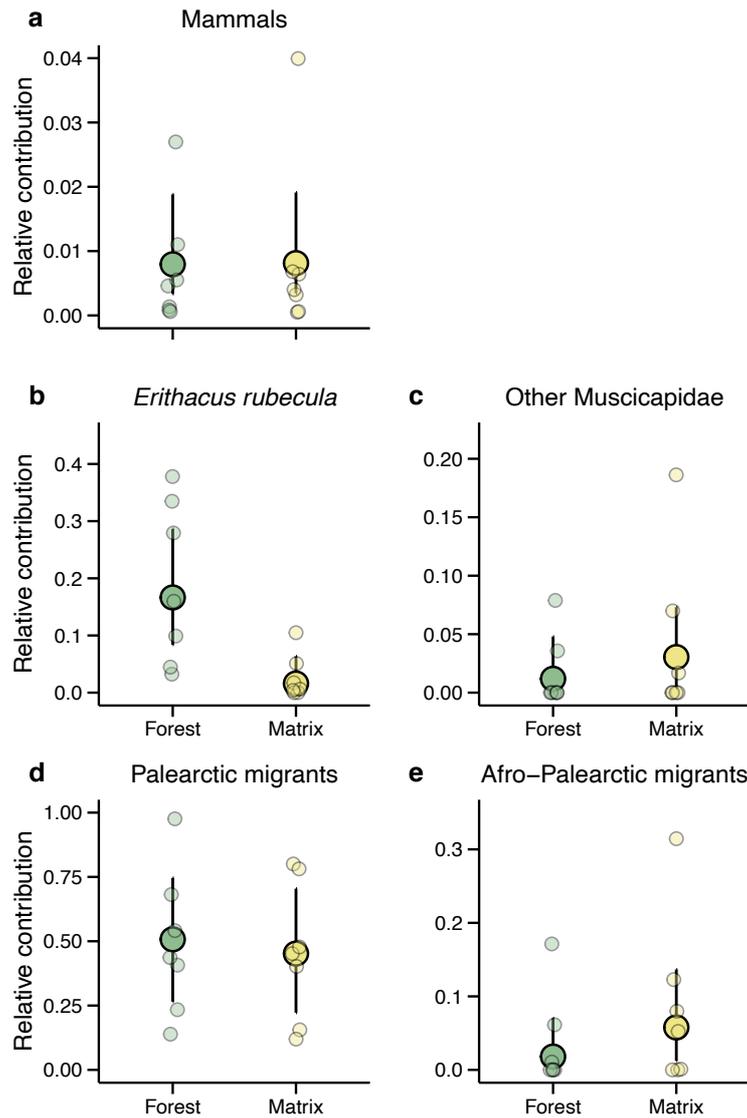


Figure S7. Differences between forest and matrix in the mean relative contribution to community-wide seed rain of (a) mammals (~ 0.008 in both habitats; $\chi^2 < 0.01$, $P = 0.960$); (b) *Erithacus rubecula* (0.167 vs 0.016; $\chi^2 = 74.50$, $P < 2 \times 10^{-16}$); (c) all Muscicapidae species but *E. rubecula* (0.047 vs 0.064; $\chi^2 = 2.52$, $P = 0.113$); (d) frugivorous birds that are Palearctic migrants (0.507 vs 0.452; $\chi^2 = 0.476$, $P = 0.490$); and (e) frugivorous birds that are Afro-Palearctic migrants (0.053 vs 0.089; $\chi^2 = 15.50$, $P = 10^{-4}$). Large circles and bars denote back-transformed means \pm 95% confidence intervals estimated by GLMMs with beta distribution and logit link ('landscape' included as random factor), whereas small circles denote observed values. Panels 'b' and 'c' show that results of Muscicapidae reported in Fig. 5c were largely driven by a single species (*E. rubecula*), as the contribution of the other six Muscicapidae species (see Table S3) was evenly distributed between habitats. Panels 'd' and 'e' show that results of the percentage of migrant frugivores reported in Fig. 6c were largely driven by Palearctic migrants, as the contribution of Afro-Palearctic migrants was smaller but higher in matrix than in forest. Palearctic migrants are birds that breed in Europe and winter in southern Europe and Africa north of the Sahara). Afro-Palearctic migrants are birds that breed in Europe and winter in sub-Saharan Africa.

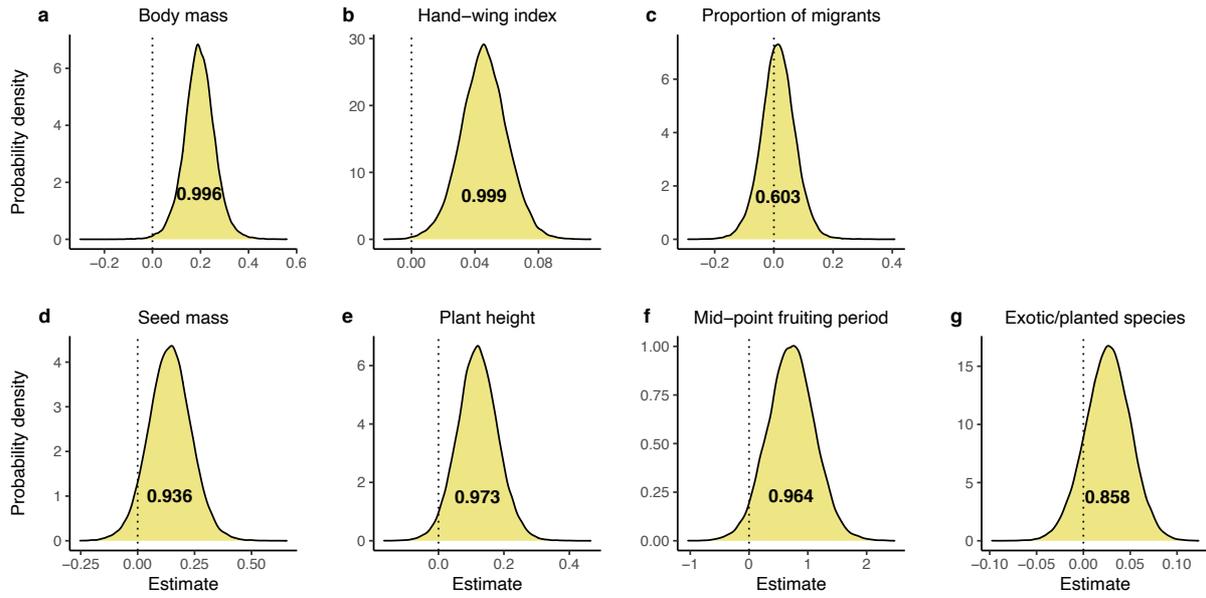


Figure S8. Posterior distribution of estimates for the ‘matrix’ effect (intercept = ‘forest’) on community-weighted mean (CWM) of frugivore (a-c) and (d-g) plant traits estimated by two generalized joint attribute models for frugivores and plants, respectively. Values in bold denote the proportion of the posterior distribution above zero (vertical dotted line). CWM body mass, seed mass and plant height were obtained –and modelled– from \log_{10} -transformed data; the hand-wing index (%) was modelled as a proportion; and the mid-point fruiting period was modelled after transforming the data as $x' = x/10$ to reduce differences in scale between plant traits (48).

References

1. I. Izhaki, P. B. Walton, U. N. Safriel, Seed shadows generated by frugivorous birds in an eastern mediterranean scrub. *J. Ecol.* **79**, 575–590 (1991).
2. K. D. Holl, Do bird perching structures elevate seed rain and seedling establishment in abandoned tropical pasture? *Restor. Ecol.* **6**, 253–261 (1998).
3. J. P. González-Varo, J. M. Arroyo, P. Jordano, Who dispersed the seeds? The use of DNA barcoding in frugivory and seed dispersal studies. *Methods Ecol. Evol.* **5**, 806–814 (2014).
4. B. Rumeu, I. Donoso, J. Rodríguez-Pérez, D. García, Frugivore species maintain their structural role in the trophic and spatial networks of seed dispersal interactions. *J. Anim. Ecol.* **89**, 2168–2180 (2020).
5. J. P. González-Varo, C. S. Carvalho, J. M. Arroyo, P. Jordano, Unravelling seed dispersal through fragmented landscapes: Frugivore species operate unevenly as mobile links. *Mol. Ecol.* **26**, 4309–4321 (2017).
6. P. Kurek, T. H. Sparks, P. Tryjanowski, Electricity pylons may be potential foci for the invasion of black cherry *Prunus serotina* in intensive farmland. *Acta Oecologica* **62**, 40–44 (2015).
7. N. M. Haddad, *et al.*, Habitat fragmentation and its lasting impact on Earth’s ecosystems. *Sci. Adv.* **1**, e1500052 (2015).
8. C. M. Herrera, Frugivory and seed dispersal by carnivorous mammals, and associated fruit characteristics, in undisturbed Mediterranean habitats. *Oikos* **55**, 250–262 (1989).
9. J. P. González-Varo, J. V. López-Bao, J. Guitián, Functional diversity among seed dispersal kernels generated by carnivorous mammals. *J. Anim. Ecol.* **82**, 562–71 (2013).
10. A. García-Rodríguez, *et al.*, Functional complementarity of seed dispersal services provided by birds and mammals in an alpine ecosystem. *J. Ecol.* **110**, 232–247 (2022).
11. P. Jordano, E. W. Schupp, Seed disperser effectiveness: the quantity component and patterns of seed rain for *Prunus mahaleb*. *Ecol. Monogr.* **70**, 591–615 (2000).
12. J. M. Fedriani, A. J. Manzaneda, Pre- and postdispersal seed predation by rodents: balance of food and safety. *Behav. Ecol.* **16**, 1018–1024 (2005).
13. J. P. González-Varo, J. M. Arroyo, P. Jordano, The timing of frugivore-mediated seed dispersal effectiveness. *Mol. Ecol.* **28**, 219–231 (2019).
14. J. P. González-Varo, *et al.*, Limited potential for bird migration to disperse plants to cooler latitudes. *Nature* **595**, 75–79 (2021).
15. M. Alcaide, *et al.*, Disentangling vector-borne transmission networks: a universal DNA barcoding method to identify vertebrate hosts from arthropod bloodmeals. *PLoS ONE* **4**, e7092 (2009).
16. S. Ratnasingham, P. D. N. Hebert, bold: The Barcode of Life Data System (<http://www.barcodinglife.org>). *Mol. Ecol. Notes* **7**, 355–364 (2007).
17. P. Torroba Balmori, P. Zaldívar García, Á. Hernández Lázaro, *Semillas de frutos carnosos del norte ibérico: Guía de identificación* (Ediciones Universidad de Valladolid, 2013).
18. P. W. G. CBOL, *et al.*, A DNA barcode for land plants. *Proc. Natl. Acad. Sci.* **106**, 12794–12797 (2009).
19. S. F. Altschul, *et al.*, Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. *Nucleic Acids Res.* **25**, 3389–3402 (1997).
20. J. P. González-Varo, S. Díaz-García, J. M. Arroyo, P. Jordano, Seed dispersal by dispersing juvenile animals: a source of functional connectivity in fragmented landscapes. *Biol. Lett.* **15**, 20190264 (2019).
21. D. M. Dehling, *et al.*, Functional relationships beyond species richness patterns: trait matching in plant–bird mutualisms across scales. *Glob. Ecol. Biogeogr.* **23**, 1085–1093 (2014).
22. M. Schleuning, J. Fründ, D. García, Predicting ecosystem functions from biodiversity and mutualistic networks: an extension of trait-based concepts to plant–animal interactions. *Ecography* **38**, 380–392 (2015).
23. M. Galetti, *et al.*, Functional extinction of birds drives rapid evolutionary changes in seed size. *Science* **340**, 1086–1090 (2013).
24. C. Sheard, *et al.*, Ecological drivers of global gradients in avian dispersal inferred from wing morphology.

- Nat. Commun.* **11**, 2463 (2020).
25. S. Claramunt, M. Hong, A. Bravo, The effect of flight efficiency on gap-crossing ability in Amazonian forest birds. *Biotropica* **54**, 860–868 (2022).
 26. J. A. Tobias, *et al.*, AVONET: morphological, ecological and geographical data for all birds. *Ecol. Lett.* **25**, 581–597 (2022).
 27. H. Wilman, *et al.*, EltonTraits 1.0: Species-level foraging attributes of the world’s birds and mammals. *Ecology* **95**, 2027–2027 (2014).
 28. I. Newton, *The migration ecology of birds* (Elsevier, 2010).
 29. J. M. Olesen, *et al.*, Missing and forbidden links in mutualistic networks. *Proc. R. Soc. Lond. B Biol. Sci.* **278**, 725–732 (2011).
 30. C. Coulson, P. G. Spooner, I. D. Lunt, S. J. Watson, J. Diez, From the matrix to roadsides and beyond: the role of isolated paddock trees as dispersal points for invasion. *Divers. Distrib.* **20**, 137–148 (2014).
 31. R. T. Corlett, Interactions between birds, fruit bats and exotic plants in urban Hong Kong, South China. *Urban Ecosyst.* **8**, 275–283 (2005).
 32. J. Tous, I. Batlle, A. Romero, Prospección de variedades de algarrobo en Andalucía. *ITEA* **91**, 164–174 (1995).
 33. P. Jordano, Data from: Angiosperm fleshy fruits and seed dispersers: a comparative analysis of adaptation and constraints in plant-animal interactions. *Dryad Data* (2013) <https://doi.org/10.5061/dryad.9tb73>.
 34. J. Kattge, *et al.*, TRY plant trait database – enhanced coverage and open access. *Glob. Change Biol.* **26**, 119–188 (2020).
 35. G. López González, *Guía de los árboles y arbustos de la Península Ibérica: (especies silvestres y las cultivadas más comunes)* (Ediciones Mundi-Prensa, 2004).
 36. S. Springmann, R. Rogers, H. Spiecker, Impact of artificial pruning on growth and secondary shoot development of wild cherry (*Prunus avium* L.). *For. Ecol. Manag.* **261**, 764–769 (2011).
 37. J. J. Camarero, Linking functional traits and climate-growth relationships in Mediterranean species through wood density. *IAWA J.* **40**, 215-S2 (2019).
 38. N. M. Fyllas, *et al.*, Functional trait variation among and within species and plant functional types in mountainous Mediterranean forests. *Front. Plant Sci.* **11** (2020).
 39. C. P. Carmona, *et al.*, Agriculture intensification reduces plant taxonomic and functional diversity across European arable systems. *Funct. Ecol.* **34**, 1448–1460 (2020).
 40. A. Guillén, P. P. Ferrer-Gallego, V. Serena, J. B. Peris, El Algarrobo (*Ceratonia siliqua* L.), importancia paisajística, económica y perspectivas de futuro. *Chron. Naturae* **7**, 45–54 (2018).
 41. A. Aparicio, A. Hampe, L. Fernández-Carrillo, R. G. Albaladejo, Fragmentation and comparative genetic structure of four mediterranean woody species: complex interactions between life history traits and the landscape context. *Divers. Distrib.* **18**, 226–235 (2012).
 42. J. P. González-Varo, Fragmentation, habitat composition and the dispersal/predation balance in interactions between the Mediterranean myrtle and avian frugivores. *Ecography* **33**, 185–197 (2010).
 43. QGIS Development Team, QGIS Geographic Information System: Open Source Geospatial Foundation Project (2022).
 44. M. Roswell, J. Dushoff, R. Winfree, A conceptual guide to measuring species diversity. *Oikos* **130**, 321–338 (2021).
 45. N. P. Chacoff, *et al.*, Evaluating sampling completeness in a desert plant–pollinator network. *J. Anim. Ecol.* **81**, 190–200 (2012).
 46. A. Chao, *et al.*, Rarefaction and extrapolation with Hill numbers: a framework for sampling and estimation in species diversity studies. *Ecol. Monogr.* **84**, 45–67 (2014).
 47. T. C. Hsieh, K. H. Ma, A. Chao, iNEXT: an R package for rarefaction and extrapolation of species diversity (Hill numbers). *Methods Ecol. Evol.* **7**, 1451–1456 (2016).
 48. J. S. Clark, Generalized Joint Attribute Modeling. Comprehensive R Archive Network (CRAN) (2016).