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Diet of Rooks *Corvus frugilegus* and potential seed dispersal in urban and agricultural habitats of Romania and Poland

Ignacy KITOWSKI[1]*, Attila D. SÁNDOR[2], Joanna CZARNECKA[3] and Grzegorz GRZYWACZEWSKI[4]

1. State School of Higher Education in Chełm, Pocztowa 54, 22-100 Chelm, Poland.
2.Department of Parasitology and Parasitic Diseases, University of Agricultural Sciences and Veterinary Medicine of Cluj-Napoca, Calea Manastur 3-5, Cluj-Napoca 400372, Romania.
3. Department of Ecology, Institute of Biology and Biochemistry, Maria Curie-Skłodowska University, Akademicka 19, 20-033 Lublin, Poland.
4. Department of Zoology, Animal Ecology and Wildlife Management, University of Life Sciences in Lublin, Akademicka 13, 20-950, Lublin, Poland.
*Corresponding author, I. Kitowski, E-mail: ignacyk@autograf.pl

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Abstract. The aim of our study was to evaluate the relationship between dispersed seed pool and the diet structure of the Rook *Corvus frugilegus* in two geographical regions of Central and Southeast Europe. We studied the content of regurgitation pellets (N=188) collected in urban and agricultural habitats of Poland and Romania at the end of the breeding period. Seeds (19 taxa) were present in 49% of analysed pellets; the frequency of pellets with seeds was higher in urban habitats in both regions. The vast majority of seeds represented fleshy-fruited species and the pulp of the fruits was an important part of the Rook's diet. The most numerous and frequent was *Cerasus avium* / *C. vulgaris*. Seeds and fruits of the dry-fruited species were present in small amounts and with low frequencies. Rooks are successful dispersers of seeds and may contribute to the diversity of vegetation especially in urban areas.

Key words: Rook, anthropogenic habitats, endozoochory, fleshy fruits, nonstandard dispersal.

Introduction

Biological diversity in Europe is highly influenced by the structure and composition of human land-use. In the past, interactions between human activities (i.e. agriculture, forestry, habitation) and nature (geomorphology, soil properties, water conditions, etc.) were expressed in the diversity of cultural landscapes. Presently, however, this diversity is being lost at an accelerated pace, while the main effects of ongoing human activity are represented by habitat fragmentation and homogenisation, which strongly affect the entire structure of the anthropogenic landscape (Jongman 2002). The problem of homogenisation is crucial for urban habitats. It was proved that urban habitats across the globe are much more uniform than the natural environment outside them (McKinney & Lockwood 1999, McKinney 2006).

The survival of plant metapopulations in such island-like fragmented landscapes depends on effective colonisation and re-colonisation of isolated favourable patches (Merriam 1988, Pulliam 1988). Habitat fragmentation and homogenisation, which lead to the reduction of favourable habitat areas and damage connectivity between patches, emphasise the importance of seed dispersal for long distances (LDD, usually defined as dispersal for distances exceeding 100 m) and of nonstandard dispersal vectors, which are not connected directly with seed morphological adaptations (Trakhtenbrot et al. 2005, Nathan et al. 2008).

Birds are some of the most effective vectors for LDD in natural and human dominated ecosystems. Our previous studies conducted in Polish human-influenced habitats proved that the Rook *Corvus frugilegus* plays an important role in the dispersal of seeds of dry- and fleshy-fruited species (Czarnecka & Kitowski 2013, Czarnecka et al. 2013).

The goal of our present research was to study the process of endozoochorous transport of seeds in urban and agricultural landscapes by analysing the relationship between dispersed seed pool during the breeding period of the Rooks and their diet structure in two geographical regions of Central and Southeast Europe divided by the Carpathian Mountains.

Material and methods

Studied species

Similarly to other corvids, the Rook is strongly connected to and highly abundant in human altered open landscapes in Europe (Tryjanowski et al. 2009). Because of this and due to its well-known biology and ecology it serves as a good model species. The Rook is an abundant species both in Poland, reaching about 250 000 - 310 000 pairs in Poland (Jakubiec 2005, Tryjanowski et al. 2009, BirdLife International 2012), with a similar abundance reported also from Romania (150 000-200 000 pairs, BirdLife International 2012). The species is an opportunistic omnivore feeding on the easiest accessible food at a particular moment (Tryjanowski et al. 2009). The diet of the Rook contains plant and animal items in equal proportion, with decreasing importance of plant material during summer when animal food is more easily accessible (Jabłoński 1979, Gromadzka 1980). Breeding colonies (rookeries) are usually located in the close vicinity of or even within human settlements in groups of trees, parks or burial grounds (Ptaszyk & Winiecki 2005). The majority of birds feed in the distance of 0.5-1 km away from the colony; meadows, pastures and spring corn are preferred, along with wasteland areas, which are visited with lower frequency (Kasprzykowski 2003). Undigested parts of food are regurgitated in the form of pellets one or two days after swallowing; the mean number of pellets formed by one bird per day ranges between 0.7 and 1.1 (Luniak 1977).

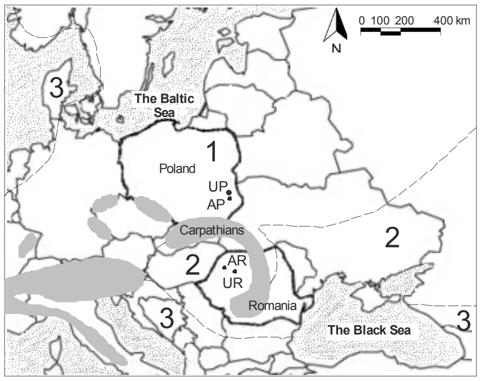

Figure 1. Distribution of the study sites (UP – urban habitat Poland, AP – agricultural habitat Poland, UR – urban habitat Romania, agricultural habitat Romania) against the background of geobotanical units in Europe according to Szafer and Zarzycki (1972). Grey color – Mountain Subprovinces, 1 – Central European Province, 2 – Pontic-Pannonian Province, 3 – other geobotanical units

Study site and methods

Regurgitation pellets were collected from 4 breeding colonies (rookeries) located in public parks in late June 2014. Two of them were located in Poland (Central European Province with the dominance of deciduous and mixed forests in natural vegetation) and two in Romania, inside of the arc of the Carpathians (Continental bioregion, Pontic-Pannonian Province, hilly areas with deciduous forests, grasslands and arable land in equal proportion, harsh continental climate, Fig. 1). One colony in each country was located in small settlements (Sielec in Poland labelled AP, colony size – 262 pairs, number of collected pellets *N*=50; Aleşd in Romania, AR, colony size – 14, *N*=50); another one in a city (Chełm in Poland, UP, colony size – 121 pairs, *N*=50; Cluj in Romania, UR, colony size – 22, *N*=38).

All pellets (one pellet = one sample) were dried, weighted and examined under the magnification 10×. All seeds were identified, and the abundance of other items helpful in the determination of the diet composition was evaluated using the following classes: 0 – absence, 1 – sporadic items (proportion < 1%), 2 – items of medium abundance (proportion 1-40%), 3 – dominant items (proportion > 40%, usually 90% or more; only when the items with medium abundance were present was their proportion was lower in the same sample). The following item classes were distinguished: (1) cereal remains including fragments of grains and husks, (2) remains of the pulp of fleshy-fruits, (3) other unidentified plant material, (4) insect fragments, and (5) rubbish (fragments of paper, plastic bags, glass, aluminium foil, bricks, etc.).

The number of seeds present in pellets was calculated per 100 pellets. The Principal Components Analysis (PCA) with the help of the MVSP software (Kovach 2005) was conducted to estimate the diet variability in the two geographic regions (Poland *vs.* Romania) and different habitat types (agricultural *vs.* urban) in relation with the seed pool present in pellets. The Statistica.Pl software was used to calculate the Spearman rank correlation (r_s) and to perform ANOVA (Sokal & Rohlf 1995).

Results

Diet analysis

The mean dry weight of pellets varied between 0.90 g (AR and UR) and 1.13 g and 1.23 g (UP and AP respectively). The main substratum of pellets consisted of different types of plant material; cereal remains were present in 39% of pellets, fleshy fruits remains in 40%, other plant material in 28%. Insect remains were a frequent admixture (mostly chitin fragments), present in 67% of samples. Cereal remains were more frequent in pellets collected in agricultural landscape (39% for AP, 70% for AR and 26% for UP and 37% for UR), reversely as in the case of fleshy fruits remains, which were more frequent in pellets collected in the cities (54% for UP and 66% for UR in comparison with 38% for AP and 12% for AR).

Results of the Principal Correspondence Analysis (abundance of diet items was taken into consideration) showed that the main factor responsible for diet structure is habitat type (agricultural *vs.* urban, Fig. 2). The Rooks whose breeding colonies were located in cities fed on fleshy fruits more often and the remains of the pulp of fleshy fruits were more abundant in their pellets. In the case of the Rooks with rookeries in agricultural landscape, fleshy fruits were less abundant and

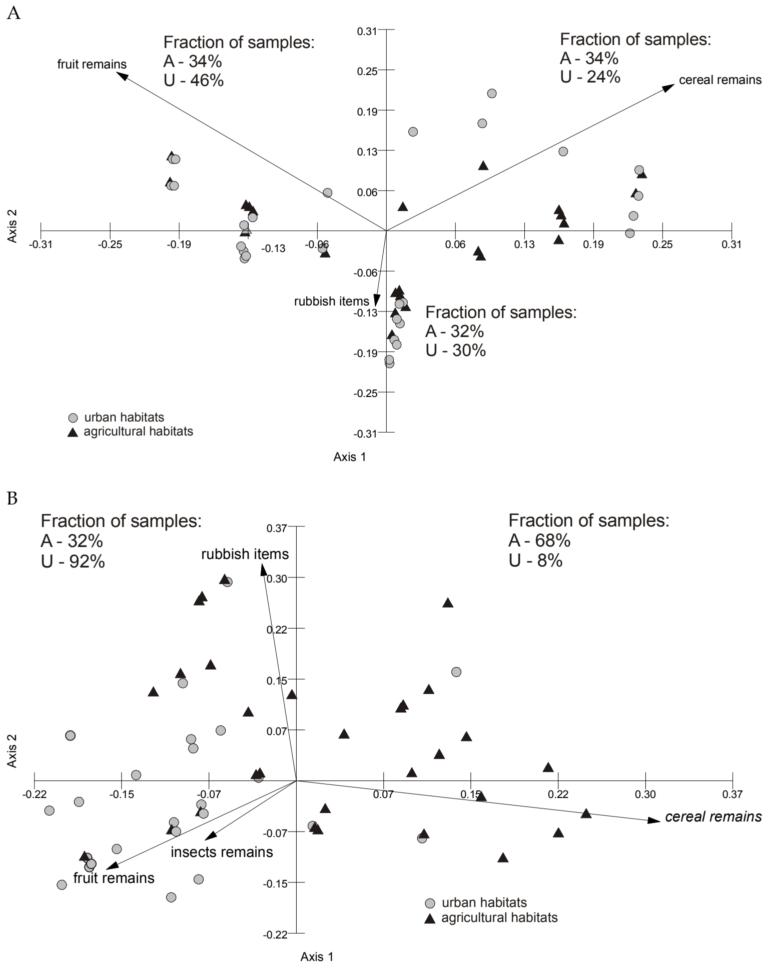

Figure 2. PCA diagram based on the presence of remains of the main diet components in pellets made for both countries separately. A – samples from Poland; eigenvalues: axis 1 – 2.067, axis 2 – 0.911; cumulative percentage variance: axis 1 – 46.23, axis 2 – 66.60. Statistically significant Spearman rank correlation coefficients (r_s) variables and axis 1: habitat type (agricultural versus urban) r_s = -0.23, $p < 0.05$; number of seeds of fleshy-fruited species r_s = -0.25, $p < 0.001$; number of cereal grains r_s = +0.34, $p < 0.001$; number of *Cerasus* sp. stones r_s = -0.24, $p < 0.05$; B – samples from Romania; eigenvalues: axis 1 – 1.959, axis 2 – 1.193; cumulative percentage variance: axis 1 – 43.64, axis 2 – 70.21. Statistically significant Spearman rank correlation coefficients (r_s) variables and axis 1: habitat type (agricultural versus urban) r_s = -0.70, $p < 0.0001$; number of *Cerasus* sp. stones r_s = -0.24, $p < 0.05$. Fraction of samples – share of samples with value of Axis 1 coordinate below zero (left side) and above zero (right side) and value of Axis 2 coordinate below zero for Polish samples (bottom part)

were partly replaced with cereal grains, whose remains were more frequent in pellets collected there. This pattern of food type replacement was much more evident for pellets collected in Romania, where the habitat type was strongly correlated with axis 1 coordinates, than it was observed for pellets collected in Poland (Fig. 2 A, B). Additionally, the presence of rubbish items was an important differentiating factor only for Polish samples. They were present in 20% of samples from agricultural (AP) and in 58% from urban habitats (UP). It was 50% and 32% respectively for agricultural and urban habitats of Romania (AR and UR).

<u>Seed pool in pellets</u>

Seeds were present in 49% of analysed pellets (*N*=188), but the frequency of pellets with seeds varied among the analysed sites: the highest was for UP (64%), and the lowest for AR (30%). The frequency of pellets with seeds of dry-fruited species was noticeably lower. It exceeded 10% only in the case of the AP habitat (Table 1).

All the seeds found in pellets represented 19 taxa; the vast majority of seeds represented fleshy-fruited species and the pulp of the fruits was an important part of the Rooks' diet (Table 1, Fig. 3). The most numerous and frequent in this group was *Cerasus avium* / *C. vulgaris*. Cherry and sweet cherry stones were present in pellets collected in all regions and habitat types. Less frequent and numerous are *Cerasus mahaleb* (a Southern European species) and *Morus alba* present only in pellets from Romania and *Fragaria* × *ananassa*, frequently planted in Poland. Cereal grains were found only in pellets collected in Poland and they were present in pellets collected in both urban and agricultural habitats. Seeds and fruits of other dry-fruited species (9 taxa) were present in small amounts and with low frequencies; the best represented taxa were *Stellaria media* (AP and UP) and *Poa annua* (UR).

The most important factor responsible for the number of seeds of fleshy fruited species and cereal grains present in pellets was the region in which pellets were collected (cf. the higher number of seeds of fleshy fruited species and the absence of cereal grains in Romania, Table 2). Habitat type was statistically important only for the abundance of *Cerasus* stones – they were significantly more abundant in cities, coinciding highly with the diet analysis results. There was no evident factor responsible for species richness and the number of seeds of dry-fruited species in pellets (Table 2).

Discussion

The role of birds, especially frugivores, in the seed dispersal process is broadly recognized (Sekercioglu 2006). Seed dispersal is a spatial process that is crucial for determining the structure and dynamics of plant populations and communities; it enables the colonisation of new habitats and helps to maintain plant diversity; it plays an important role in succession and regeneration; and it also affects the rates of gene flow and influences the genetic structure of populations (Howe & Miriti 2000, Nathan& Muller-Landau 2000, Wang & Smith 2002).

Seed dispersal can be called "specialised", when it is mediated by a standard vector to which the seeds are particularly adapted, and "generalised", i.e. mediated by a non-standard vector. Morphological dispersal adaptation and in consequence standard means of dispersal usually generate local dispersal; non-standard vectors are responsible for long-distance dispersal (LDD), which is so important in fragmented, man-shaped landscape (Higgins et al. 2003, Nathan et al. 2008, Nogales et al. 2012). Here, we confirmed the idea of Czarnecka & Kitowski (2013) that the Rook is a standard dispersal vector for fleshy-fruited species like *Cerasus* sp., *Fragaria* × *ananassa* and *Morus alba*, and simultaneously it is also a non-standard vector for less frequently distributed dry-fruited species other that cereals, which were probably unintentionally swallowed during foraging for other food items. Events of non-standard dispersal are rare and impossible to predict and their role in plant dynamics was underappreciated for a long time. Now the awareness of the importance of rare events increases significantly and it is broadly accepted that even a small proportion of seeds moving for long distances can increase a predicted plant spread rate (Wilkinson 1997, 1999, Clark et al. 1998, Higgins & Richardson 1999, Higgins et al. 2003, Nathan et al. 2003, Nogales et al. 2012).

Omniviorous birds, with gulls and corvids among them, obtaining food from a wide range of resources, are thought to have a great capacity for seed transportation between oceanic islands (Nogales et al. 2012). Seed dispersal in man-shaped landscape faces the same problems as their transport between oceanic islands (MacArthur & Wilson 1967) and the same mechanisms guaranteeing long-distance dispersal are important there. Dynamic agricultural landscape is often described successfully with the help of the "patch-matrix" model, where small areas favourable for plant vegetation (patches) are "submerged" in unfavourable environment of the "matrix" (Marshall 2002). The Rook is one of the important dispersal vectors linking favourable patches surrounded by the unfavourable matrix (arable fields in agricultural landscape or highly transformed areas in urban landscape) and is able to transport some amounts of seeds without any external adaptations to endozoochory (see the list of species in: Czarnecka & Kitowski 2010, 2013, Czarnecka et al. 2013).

Two aspects of seed dispersal involving birds must be taken into consideration: "vectors in spread", which affect the transport of propagules

Table 1. Composition of the seed pool in pellets; AP – agricultural habitat in Poland, UP – urban habitat in Poland, AR– agricultural habitat in Romania, UR – urban habitat in Romania. Nomenclature according to Mirek et al. (2002)

Taxa	Number of seeds per 100 pellets				Frequency (%)			
	AP	UP	AR	UR	AP	UP	AR	UR
Fleshy-fruited species								
Cerasus avium (L.) Moench/*C. vulgaris* Mill.	86	134	64	150	30	48	32	47
Cerasus mahaleb (L.) Mill.			40				18	
Fragaria × ananassa Duchesne	6	18			2	4		
Morus alba L.			62	87			14	16
Padus avium Mill.				29				3
Cereals and other cultivated dry-fruited species								
Avena sativa L.	2				2			
Hordeum vulgare L.	20	2			10	2		
Linum usitatissimum L.	4	4			2	2		
Triticum aestivum L.	36	30			6	6		
Zea mays L.	4			5	4			3
Dry-fruited species								
Avena fatua L.	2				2			
Echinochloa crus-galli (L.) P. Beauv.		2				2		
Galium aparine L.		2				2		
Poa annua L.				21				3
Polygonum aviculare L.				3				3
Robinia pseudoacacia L.	2				2			
Stellaria media (L.) Vill.	26	20			8	4		
Trifolium dubium Sibith.			2				2	
Veronica hederifolia L.	2				2			
Total number of seeds	190	212	168	295	-	-	-	-
Total number of taxa	11	8	4	6	-	-	-	-
Pellets with seeds	-	-	-	-	44	64	30	63
Pellets with seeds of dry-fruited species	-	-	-	-	14	8	2	5

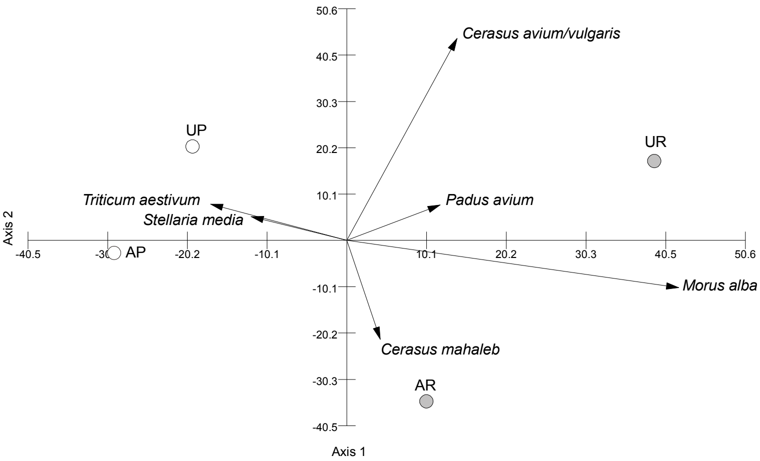

Figure 3. PCA diagram based on the presence of seeds in samples. Eigenvalues: axis 1 – 2880.12, axis 2 – 1962.80; cumulative percentage variance: axis 1 – 57.20, axis 2 – 96.20.

discussed above, and "agencies of establishment", which affect local conditions in sites were seeds were placed (Chambers 1999). Breeding colonies of the Rooks (rookeries) are formed in places with a me-dium level of man-made disturbances (Petrescu 2002, Jakubiec 2005, Orłowski & Czapulak 2007, Gache 2014), and thus with small vegetation gaps that enable the germination of transported seeds (Czarnecka & Kitowski 2010). Additionally, the presence of the Rook colonies alters soil conditions, thus contributing to the increase in soil humidity and content of biogenic elements,

Table 2. Results of two-way ANOVA of the differences in species pool of pellets (N=188) between samples from different regions (Poland *vs*. Romania) and between different habitat types (agricultural *vs.* urban); $^*p$ < 0.05, $^{**}p$ < 0.01

Variable	Region (df = 1)		Habitat type (df = 1)		Region x habitat type (df = 1)	
	F	p	F	p	F	p
Species richness	0.00	0.989	0.12	0.730	0.12	0.730
Number of						
All seeds	0.33	0.564	2.00	0.159	0.99	0.320
Seeds of fleshy-fruited species	4.89	0.028*	3.54	0.061	0.22	0.640
Cerasus stones	0.01	0.922	4.81	0.030*	0.39	0.535
Seeds of dry-fruited species	0.80	0.373	0.16	0.687	0.76	0.383
Cereal grains	8.50	0.004**	0.56	0.457	1.13	0.289

which are also helpful during seed germination and seedling establishment (Borkowska et. al. 2015).

The frequency of occurrence of the main components of the birds' diet – fleshy fruits, cereal grains and food found in human refuse (we can suspect it because of the presence of different rubbish items like paper, plastic and aluminium foil in pellets) – was not strongly influenced by the geographic region and phytogeographical province (Central European *vs.* Continental Province) where the pellets were collected. The most important factor was the type of main crops grown, which are the basic food resources for the Rooks. Food availability and the diversity of food resources determine the number and species structure of dispersed seeds; such a correlation was previously demonstrated for the Grey Partridge *Perdix perdix* (Orłowski & Czarnecka 2013). For example, higher accessibility of *Prunus mahaleb* and *Morus alba* fruits in Romania was responsible for observed differences in diet – both species are connected with warmer and drier habitats and in consequence they are more abundant in Romania than in Poland. Further studies of the interaction between food resources and seed dispersal mediated by the Rook, a bird species with a broad geographical range, in different phytogeographical regions may bring more information about the diet and seed dispersal pattern.

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