RESEARCH ARTICLE



Diet analysis and the assessment of plastic and other indigestible anthropogenic litter in the white stork pellets

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Abstract

Pollution by anthropogenic litter is a major threat to global ecosystems. Seabirds are frequently used as environmental monitors of litter ingestion, but similar research is rare for terrestrial birds. Here, we focused on pellet analysis from 117 nests of an iconic bird of the Western Palearctic, the white stork (*Ciconia ciconia*), breeding in southern and southwestern Poland in a farmland landscape, far away from large dumps and landfills. We found that most prey items in the diet of white storks were invertebrates (particularly from orders Coleoptera, Orthoptera, and Hymenoptera) but vertebrate prey comprised most of the biomass. Further analysis revealed that anthropogenic litter was found in 22.7% of pellets (34.2% of breeding pairs) with plastic (8.4%) and cigarette filters (6.9%) being most prevalent. This study represents the first assessment through pellet analysis of the ingestion of anthropogenic litter by live wild storks in Poland and also by a migratory population of white storks. Our study indicates a potentially significant transfer of plastic and other anthropogenic material through terrestrial food webs.

Keywords Anthropogenic litter · Biological indicators · Diet composition · Microplastic · Pellet analysis · Terrestrial ecosystems

Introduction

Anthropogenic litter is a widespread form of pollution that is a major threat to global ecosystems (Bergmann et al. 2015; Deudero and Alomar 2015; Kaza et al. 2018; MacLeod et al. 2021). In 2020, 2.24 billion tonnes of solid waste was generated, and annual waste production is expected to increase to almost 4 billion tonnes in 2050 (Kaza et al. 2018). More than 50% of seabird species are affected by marine anthropogenic (particularly plastic) litter, either via ingestion or entanglement (Kühn et al. 2015; Wilcox et al. 2015), and it has been predicted that plastic ingestion will affect virtually all seabird

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species by 2050 (Wilcox et al. 2015). Hence, seabirds have often been used as biological monitors of anthropogenic litter in the marine environment (e.g., Acampora et al. 2017; Mallory et al. 2010; Ryan 1987a; van Franeker and Law 2015). Seabirds may mistake indigestible anthropogenic litter for prey or ingest it with real prey. The harmful effects of ingestion of plastic and other litter may involve a reduction in digestive capacity (Furness 1985a; Pierce et al. 2004; Ryan 1988) and physical damage through perforation of and blockage of the digestive tract causing ulcerations (Bourne and Imber 1982; Pettit et al. 1981; Pierce et al. 2004), leading to reduction of nutrition intake and starvation (Pierce et al. 2004) and

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reduced body mass (Lavers et al. 2014; Pierce et al. 2004; Ryan 1987b; Spear et al. 1995) and growth (Danner et al. 2011; Ryan 1988). Combined with the potential chemotoxicity of the litter (Tanaka et al. 2013), these may contribute to reduced survival and reproduction in animals (Bouland et al. 2012; Pierce et al. 2004; Roman et al. 2019; Ziccardi et al. 2016).

Litter in the diet of seabirds is commonly monitored through analysis of the stomach content of dead birds, where indigestible material is typically accumulated (Codina-García et al. 2013; Ryan 1987a; van Franeker et al. 2011; van Franeker and Law 2015). This is particularly the case for groups such as Procellariiformes, which are unable to regurgitate indigestible material in the form of pellets and boluses (Azzarello and van Vleet 1987; Furness 1985a). Thus, ingested litter may accumulate in their gastro-intestinal system for long periods before being eliminated (Furness 1985b). However, several other groups of seabirds, and also terrestrial birds, can regurgitate undigested food in the form of pellets and these are frequently used as a data source in studies of the avian diet (Barrett et al. 2007; Speakman 1991). The monitoring of live birds through pellets is a non-invasive, low-effort, and low-disturbance sampling method providing valuable and complementary information on anthropogenic litter ingestion in species that possess the ability to regurgitate indigestible material (Acampora et al. 2017; Bond et al. 2021).

Litter is widespread in terrestrial and freshwater ecosystems but the research on this topic is limited compared to that in marine ecosystems (Duis and Coors 2016; Holland et al. 2016; Horton et al. 2017; but see de Souza Machado et al. 2018). Consequently, the exposure and ingestion rates of anthropogenic litter by terrestrial animals are not well understood (Henry et al. 2011; Sherlock et al. 2022; Zhao et al. 2016). But the problem may be serious; for example, Sherlock et al. (2022) found microplastic particles in more than 80% of chicks of aerialfeeding tree swallows (*Tachycineta bicolor*). Similarly, anthropogenic litter was found in digestive tracts of 16 out of 17 (94%) of examined specimens of terrestrial birds in Shanghai, China (Zhao et al. 2016), suggesting that exposure and ingestion of anthropogenic litter may also be high in terrestrial organisms.

The white stork (*Ciconia ciconia*) is an iconic and charismatic farmland bird species of the Western Palearctic that often lives in close association with people (Elliott et al. 2020). During the breeding season, the reproductive success of white storks is strongly dependent on the food supply near their breeding sites (feeding range is typically <2 km from the nest; Denac 2006; Nowakowski 2003), with food availability consequently also affecting population dynamics at wider geographic scales (Massemin-Challet et al. 2006; Tryjanowski et al. 2005; Tryjanowski and Kuzniak 2002). White stork feeding is highly opportunistic, using whatever large invertebrates and small vertebrates are available (Elliott et al. 2020). Recently, storks have also increasingly been reported to feed at rubbish dumps, during both breeding and non-breeding seasons (Ciach and Kruszyk 2010; Tortosa et al. 2002). These novel anthropogenic sources may be dangerous for storks because animals may ingest organic litter and also pieces of indigestible anthropogenic litter, such as plastic and textiles. Indeed, plastic was found in the stomachs of 14% of adult storks and 41% of juvenile storks feeding in urban refuse dumps in Spain (Peris 2003).

Here, we collected and analyzed pellets from 117 pairs of white storks breeding in southern and southwestern Poland to quantify their exposure to indigestible anthropogenic litter debris. Firstly, we analyzed the occurrence of natural prey items (invertebrates and vertebrates) and estimated their biomass in the pellets. We then counted the number of anthropogenic items (e.g., plastic or glass) in the pellets and quantified their occurrence and prevalence in pellets. This study represents the first attempt to estimate the rate of ingestion of anthropogenic litter debris in living adult white storks through pellet analysis in Poland. This may also represent the first such assessment for migratory population of white storks because previous studies focused mainly on Iberian Peninsula where storks are part of western population and mainly sedentary (Peris 2003; Rabaça et al. 2021). Our results may help in conservation of this species and other species with a similar ecology.

Material and methods

Study area

The pellets were collected from white stork nests during mid-June and early July 2020 in southern and southwestern Poland (Fig. A1) during the late part of the breeding season. Adult and nestling pellets typically differ in their size and structure with adults generally producing larger and more stable pellets (see also Rosin and Kwiecinski 2011). We collected only large pellets which were presumably produced by adult birds. We used a lifting platform to reach nests where we collected only fresh pellets positioned on the top of each nest. Whenever possible, for each nest/breeding pair (N=117), we collected two pellets (N=85 nests); however, we found only one pellet at 32 nests. These nests were typically in farmland (agricultural) landscape, all far away (>10 km) from large refuse dumps and landfills; white storks typically forage within a radius of 5 km from their nests (Johst et al. 2001). The Polish population of the white stork has been declining since the 1950s (Wuczyński et al. 2021).

Pellet analysis

We weighed pellets, then soaked them in water, and separated material by hand in a Petri dish before they were screened through a 1-mm mesh sieve and dried (Orłowski et al. 2014).

We identified invertebrates using a binocular microscope at $\times 40$ magnification; most invertebrate remains were represented by fragments > 1 mm but we also examined smaller fragments. The number of prey individuals representing particular taxa in each pellet was estimated based on the number of chitinous body parts, particularly the elytra (for Coleoptera, Homoptera, and Heteroptera), wings (for Diptera and Hymenoptera), mouthparts (most groups), and other preserved body parts (e.g., limbs, clypeus, mandible). If we found two or more different chitin parts (e.g., mandibles or legs) belonging to the same species, we counted this as a single individual of a particular taxon, following the procedure adopted in previous studies on the diet of birds (Orłowski and Karg 2011, 2013). Vertebrate prey taxa and the number of individuals were estimated based mostly on bones (e.g., skull and limb bones), hair, nails, and scales. In most cases, we assigned same-species remains to a single prey individual in each pellet (Resano-Mayor et al. 2014). However, if we found unequivocal evidence for the presence for multiple individuals of the same taxon in a single pellet (e.g., same two parts of a mandible), we counted them separately. Whenever possible, we identified both invertebrate and vertebrate prey to species level by using the collections from the Institute of Agricultural and Forest Environment Poznań-Turew (invertebrates) and Mammal Research Institute PAS Białowieża (mammals) for reference. However, prey types differ in their digestibility and prey biomass rather number may better reflect the diet (Rosin and Kwiecinski 2011). To estimate the entire biomass of each prey item type, we estimated a dry weight (in milligrams) for each individual prey item; the dry weight of invertebrates was taken from detailed measurements based on the analysis of nearly half a million individuals of various invertebrate taxa (Karg 1989). For vertebrates, we first estimated the wet weight based on data collected in the Mammal Research Institute (mammals) or from literature sources (other vertebrates) and then we converted this to dry weight by multiplying wet weight by a coefficient of 0.27 for reptiles (Vitt 1978), 0.41 for birds (Shoffner and Brittingham 2013), and 0.32 for mammals (Baker et al. 1993). Regarding anthropogenic material in pellets, we collected information on the number of all items of this origin > 1 mm in size. We tested whether the presence of anthropogenic material (binary variable: yes or no) was correlated between the two pellets from the same stork pair using a chi-square test.

Results

Natural diet items in pellets

Detailed pellet analysis revealed that most prey items in white stork pellets included invertebrates, particularly from the orders Coleoptera (11173 individuals; 54.5% prey items), Orthoptera (2466 and 22.1%, respectively), and Hymenoptera (1707 and 15.3%, respectively) (Fig. 1). However, the dry mass of vertebrates in pellets was estimated to be 3.5 times higher than the mass of invertebrates (1759 and 507 g, respectively) (Fig. 1). For details on prey species composition, see Table A1.

Anthropogenic litter in pellets

In total, we found 194 items of anthropogenic origin in the pellets. Anthropogenic litter items were found in 22.7% of pellets and in pellets from 34.2% of pairs (Fig. 2). The

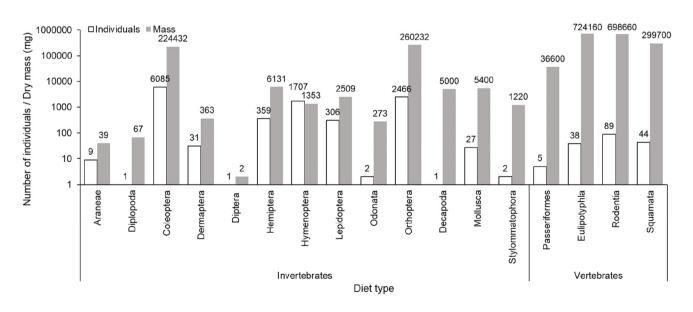


Fig. 1 Number of individuals and dry mass (mg) (both on a log scale) of invertebrate and vertebrate prey items found in white stork pellets (N=202) in Poland

most prevalent items were plastic (8.4% pellets; 21 items), cigarette filters (6.9% and 16, respectively), and glass (6.4% and 136, respectively) (Fig. 2). For details on anthropogenic items in the pellets, see Table A2. We found that the presence of anthropogenic material was correlated between the two pellets from the same pair ($\chi^2 = 17.414$, df = 1, p < 0.001).

Discussion

We found a relatively high prevalence of anthropogenic litter in the pellets of breeding white storks, a large terrestrial bird species which feeds opportunistically on various groups of invertebrates and vertebrates, in farmlands of southern and southwestern Poland. We speculate that the high occurrence of relatively small natural food items in its diet and high diet generalism makes this species particularly vulnerable to ingestion of small anthropogenic litter items. Our results indicate that anthropogenic litter is frequently ingested by some large terrestrial birds, indicating potentially significant transfer of plastic and other anthropogenic material particles through terrestrial food webs and ecosystems. To the best of our knowledge, this is the first assessment of ingestion of indigestible anthropogenic litter by live wild storks through pellet analysis in Poland. Moreover, our study may represent the first such assessment from the migratory population of white storks.

Our pellet analysis revealed that at least one item of anthropogenic origin was found in pellets from 34% of monitored white stork pairs. We also found that 23% of all pellets contained items of anthropogenic origin; the most prevalent were plastic, cigarette filters, and glass items, each category being present in >6% of pellets. Previous analyses of white stork pellets during the non-breeding season in Bulgaria also found anthropogenic litter, though with a much lower frequency of occurrence-2.7% and 4.1% of pellets for glass and plastic, respectively (Milchev et al. 2013). Similarly, Acampora et al. (2017) found that only 3.2% of pellets from great cormorants (Phalacrocorax carbo), an aquatic bird up to 1 m long, contained plastic litter. In contrast, Henry et al. 2011 found that 26% of necropsied white storks from France had rubber bands in their digestive tract and that the prevalence of anthropogenic litter in birds differed markedly between stork populations. The relatively high prevalence of anthropogenic material in French storks might be caused by the frequent occurrence of refuse dumps in the study (Henry et al. 2011) but this may also point to methodological issues (i.e., necropsy study vs. pellet analysis). Pellet analysis could underestimate the exposure of white storks to the ingestion of indigestible anthropogenic litter because not all these items were regurgitated in the form of pellets and because of the generally high digestive efficiency in white storks (Rosin and Kwiecinski 2011). Hence, some caution is needed when comparing our results with studies using other methods. However, the combined analysis of regurgitants and digestive tract content

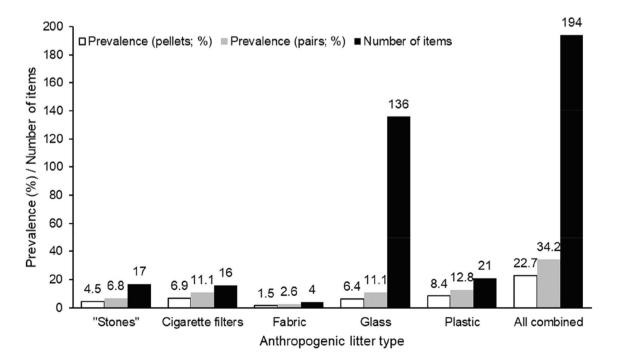


Fig.2 The prevalence (%) per pellet (N=202) and pair (N=117) and the number of indigestible anthropogenic litter items in pellets of white storks in Poland. The prevalence per pellet and pair was esti-

mated as the proportion of pellets and pairs which pellets contained at least one item of anthropogenic origin

could provide valuable additional information on the occurrence of anthropogenic litter in the diet of terrestrial birds.

Storks regurgitate pellets daily (Rosin and Kwiecinski 2011). This indicates that pellets are formed and regurgitated hours after ingestion of prey and that analysis of pellet content reflects short-term variation in composition of the diet of this species. Adult storks feed their nestlings by regurgitating food that may have indigestible items. Hence, young storks may show increased susceptibility to ingestion of anthropogenic litter items because of their lesser ability than adult birds at discriminating and regurgitating non-edible items (Henry et al. 2011; Peris 2003). We collected pellets under stork nests; hence, the collected samples might reflect both the diet of nestlings and that of adult birds if we assume that nestlings do not select specific parts of the regurgitated food supplied by adults. Our knowledge on this topic would benefit from comparison of pellet content between the breeding and non-breeding seasons as well as direct comparison of pellets regurgitated by adults and nestlings.

Conclusions

In conclusion, we showed that anthropogenic litter was frequently found in pellets of white storks. This has several practical implications. First, pellet analysis is a non-invasive method to explore and identify anthropogenic litter ingested by birds. However, future research should explore whether alternative methods, such as analysis of faeces or gastro-intestinal tracts, provide quantitatively and qualitatively similar results for tracking the circulation of anthropogenic particles in the bodies of birds. Second, it seems that large species such as white storks may be appropriate for monitoring of exposure of terrestrial wildlife to micro- but also macro-particles of anthropogenic origin, with the latter being probably more rarely documented in smaller species. The challenge for future studies lies in the investigation of potential sources of different types of anthropogenic litter in the environment, exposure of wildlife to them, concentrations and passage times of these materials through various groups of animals, and their effects on wildlife fitness.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s11356-023-31710-2.

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Author contribution LJ, KW, and PT established the original idea of the study in the field; PM with help from PT did analyses; PM wrote the first version of the manuscript with the help from PT; JK identified the prey from pellets; LJ, KW, JS, SC, KM, MP, HS, and JW collected pellets in the field. All authors accepted the final version of the manuscript. **Data availability** The data that support the findings of this study are available in the Supplementary material.

Declarations

Ethical approval and consent to participate Not applicable.

Consent for publication All authors have consent for publication.

Competing interests The authors declare no competing interests.

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