Marine Pollution Bulletin 205 (2024) 116695

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

Baseline

Blue nests: The use of plastics in the nests of the crested oropendola (*Psarocolius decumanus*) on the Brazilian Amazon coast

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A R T I C L E I N F O

Keywords:
Icteridae
Nesting
Plastic debris
Marine litter
Fishing gear
Ghost fishing
Salinópolis

A B S T R A C T

Birds have been impacted by plastic pollution via entanglement, accidental ingestion, and exposure to chemical contaminants. However, plastics were also observed as a nesting material for several species. For the first time, we describe the occurrence and composition of plastics in 36 nests of the crested oropendola (*Psarocolius decumanus*) in three different sites on the Amazon coast. Plastics were present in 67 % of abandoned, fallen nests. At the mangrove site, all nests contained plastics, while at the grassy clearing and the fishermen's village, plastics were present at 35.3 and 90 % of the nests, respectively. Blue fibers and ropes were the main plastics observed, probably derived from discarded fishing gear. Of 79 analyzed fibers, 97.5 % were composed of Polyethylene, and 83.5 % contained Cobalt Phthalocyanine as an additive. Our results showed a widespread use of plastics by *P. decumanus*, which may increase the exposure of both juveniles and adults to potential contaminants.

The Brazilian Amazon coast is a highly dynamic ecosystem, where multiple interactions occur between the great Amazon delta, the Atlantic coast, and the adjacent sea (Nittrouer et al., 2021). A myriad of ecosystems coexist in the area, like sandy beaches, rocky outcrops, vegetated dune-beach ridges, mangroves, estuarine channels, and muddy flats (Pedrini et al., 2021). The biota from the Amazon coast is also diversified, where many residential and migratory birds are benefited by the provided ecosystem services (Owuor et al., 2024).

The region bears a diversity of both marine and continental birds (Lees et al., 2014). Such animals are probably already impacted by plastic pollution, since plastic debris and microplastics are widely distributed in the Amazon biome and, particularly on the Amazon coast (Morais et al., 2024), posing a potential threat to the local biota (Pegado et al., 2018; Morais et al., 2020).

Recently, the first official record of synthetic fibers in the structure of nests of the crested oropendola, *Psarocolius decumanus* (Pallas, 1769), was published by Hoyos et al. (2021). *P. decumanus* occurs from the south of Central America to South America, except in Chile and Uruguay (e.g., Ridgely and Tudor, 2009). The bird inhabits humid forests, mangroves, dry forests, clearings, and agricultural areas (Van Perlo, 2009). Despite the least concerning status, geographic distribution, conservation, and population dynamics in Brazil are controversial and demand a new synthesis (IUCN, 2020; SIBBr, 2020). In the Brazilian Amazon, studies on *P. decumanus* are limited to checklists and geographic distribution (Sanaiotti and Cintra, 2001; Lees et al., 2014) or a few biological and ecological aspects like the mapping of nesting sites (Lopes et al., 2023) and the description of the entomological fauna associated with the nests (Torres, 2001).

Psarocolius decumanus is a tropical icterid bird, with a dark brown color with a yellow tail and yellowish-white beak; the adults measure between 35 and 48 cm (Van Perlo, 2009). The bird is known for its elaborate, long nests, which are suspended from palm leaves or branches of trees and composed of natural fibers from dry leaves, orchid roots, fungi elongated rhizomorphs (Sick, 1984), and even plastics (Hoyos et al., 2021).

Several birds often incorporate synthetic materials into their nests, such as plastic fibers and ropes (Jagiello et al., 2018), a frequently observed behavior in many seabird species (Lavers et al., 2013; Tavares

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https://doi.org/10.1016/j.marpolbul.2024.116695
Received 9 June 2024; Received in revised form 1 July 2024; Accepted 3 July 2024
Available online 11 July 2024

et al., 2016; O'Hanlon et al., 2019; Delgado et al., 2020; among others), but still poorly described for birds with a wider distribution towards coastal and continental areas (Gallitelli et al., 2023) like *P. decumanus*.

It is suggested that the use of anthropogenic materials in the composition of nests can lead to greater predation, as these nests are more easily detected (Møller, 2017; Corrales-Moya et al., 2023). Additionally, synthetic materials can cause entanglement and death of birds (Blettler and Mitchell, 2021), and they have other detrimental effects on the organism and its offspring (e.g., Teuten et al., 2009). Moreover, the presence of toxic plastic additives and adsorbed contaminants in these materials poses a significant health risk since such contaminants were reported in eggs from seabirds (van der Schyff et al., 2021).

The associated contaminants in plastics used during nesting or ingested by birds is yet rarely explored, as well as the impacts on birds' physiology and behavior (Azevedo-Santos et al., 2021). Here, we evaluated for the first time in the scientific literature the types, chemical composition and frequency of plastics and associated dyes from 36 abandoned, fallen nests of the crested oropendola, *Psarocolius decumanus* (Passeriformes: Icteridae) distributed at three sites on the Brazilian Amazon coast during field observations between 2022 and 2023. We also analyzed the chemical composition of 79 fibers to avail the composition and the presence of potential chemical contaminants (organic dyes) in these materials.

The area is located on the Atlantic coast of Pará state, which has a strong seasonality marked by the pluviosity regime and the Amazon River plume variability throughout the year. The heterogeneous coast is composed of sandy beaches, rocky outcrops, dunes, and typical coastal vegetation locally described as "restinga," mangroves, tidal channels, and muddy flats (Pedrini et al., 2021).

This study was stimulated by a first observation of plastics in a nest of the crested oropendola at Salinópolis municipality, in November 2021. In the following years, sampling took place at three distinct sites from the Pará state coast, Brazilian Amazonia (Fig. 1). The first is a mangrove patch associated with the Maçarico sandy beach at Salinópolis municipality. The typical mangrove trees were *Avicennia germinans* (L.) L. and *Laguncularia racemosa* (L.) C.F. Gaertn, where discarded fishing nets accumulated on roots (Fig. 2). The second area was a modified grassy clearing with some vacation houses and the presence of palm trees. The area is only a few meters from dune-beach ridges associated with the Farol Velho beach, also located at Salinópolis.

Finally, a fishermen village located at Maiandeua island, inside the Algodoal-Maiandeua Environmental Protection Area (municipality of Maracanã) was sampled. The traditional village has a low density of civil constructions, most of them with vegetated gardens. Mangrove and vegetated dunes are also present around the village. Abandoned and fallen nests of *P. decumanus* were collected by active search in each sampling site. Field sampling and observations were conducted between September 2022 and September 2023. Only abandoned, discarded nests were sampled, individually placed in boxes, and transported to the laboratory.

At the laboratory, each nest was visually inspected for the detection of plastic debris. The presence of plastics was evident in some of the nests due to the large number of plastic fibers and the blue color, thus named as blue nests (Fig. 2). Since the number of plastics was small for some of the nests, all were inspected by completely separating the potential plastic litter from the organic matter. The potential plastic litter composed exclusively by fibers and ropes were separated from the organic matter, with the aid of a stereomicroscope when necessary. Particles below 5 mm in length (microplastics) were not analyzed. The isolated plastics were then classified by type according to GESAMP (2019) and by color according to UNEP (2020).

To confirm the polymeric nature of the potential plastic fibers, 79 filaments from the three different sites were removed from the nests and analyzed by RAMAN spectroscopy: 16.5 % from Farol Velho, 44.3 % from the fishermen's village, and 39.2 % from the mangrove patch. The labRAM HR Evolution equipment (HORIBA) was used with lasers of different wavelengths (473, 532, 633, and 785 nm) and a 50× long-range objective (NA = 0.55). The resistance of each material was previously tested to obtain the best signal for the spectra without damaging the material. For the final spectrum measurement, the spectral region from 200 to 3200 cm^{-1} was used, allowing observation of the common polymer fingerprint region and the C-H stretching region.

Fig. 1. The three sampling sites for abandoned nests of the crested oropendola (*Psarocolius decumanus*) at Pará state, Brazilian Amazon coast. F: fishermen village (Algodoal-Maiandeua Marine Protected Area); M: mangrove area, associated to Maçarico beach; G: grassy clearings, located nearby Farol Velho beach. Map created with Datawrapper.

Fig. 2. *Psarocolius decumanus* (A); An active blue nest (B); *P. decumanus* and nesting activity with incorporation of plastic fibers (C); A piece of entangled, discarded fishing net in the border of the mangrove patch by Maçarico beach (D). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

After optimizing parameters (e.g., integration time, number of accumulations, and slit diameter), it was ensured that there was no detector saturation across the entire spectrum collection region, and the signals were typically collected with a total time between one and five minutes. A filter was used to automatically identify and remove any spikes due to cosmic rays by comparing the spectra from different accumulations (adapted from Choquette et al., 2007; Matousek et al., 2005). A baseline was applied, and a noise filter was used through the

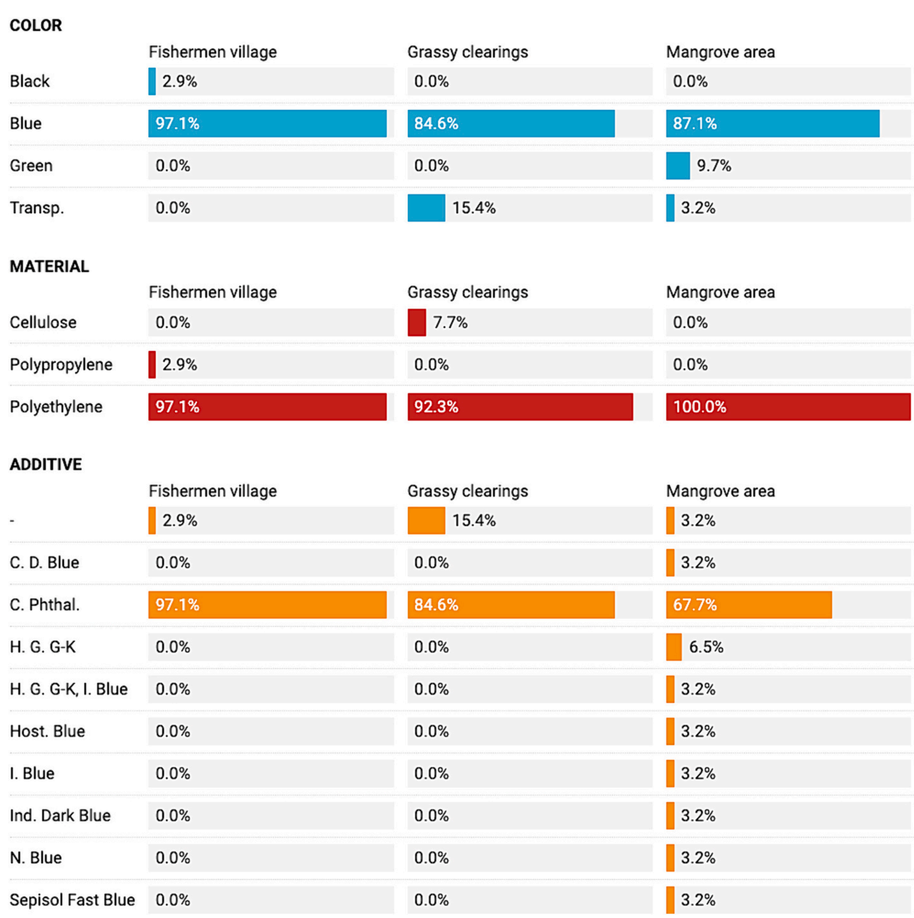

Fig. 3. Color, material (polymeric composition), and additives for the 79 analyzed fibers removed from nests of the crested oropendola (*Psarocolius decumanus*). Transp: transparent; C. Phthal.: Cobalt Phthalocyanine; C. D. Blue: Cibanom dark blue; H. G. G-K: Hostasol green G-K; I. Blue: Irgazin blue; Host. Blue: Hostasol Blue; Ind. Dark Blue: Indanthren dark blue; N. blue: Neozapon blue. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article). Figure created with Datawrapper.

micro-Raman software tools (Labspec) or a Matlab® code. The compounds in the spectra were identified using the Knowitall® program's database (Wiley, 2024).

Plastics were observed in 24 of the 36 analyzed nests. All nests from the mangrove area contained plastics ($n = 9$), classified as fibers, tangled fibers, and ropes. Blue was the dominant color, followed by green, black, and transparent fibers. For the grassy clearing, six of 17 nests contained plastics, and all of them were blue or black fibers on tangled fibers. At the fishermen's village, plastics were present in nine of 10 nests. Fibers, tangled fibers, and ropes were present in the nests. Blue, followed by green were the dominant colors, with rare occurrences of black, white, and transparent fibers. Blue was always the dominant fiber color (Figs. 2, 3).

From the 79 analyzed fibers, 77 were successfully identified as polyethylene, one as polypropylene, and one as cellulose (Fig. 3). Density was determined through RAMAN spectroscopy for 46 fibers, from which 25 were made of high-density, four of intermediate density (0.945) and 17 of low-density polyethylene.

All six identified dyes were classified as organic pigments, which were detected in 75 fibers (94.9 %). Among the fibers, 72 (91.1 %) contained blue dyes and three (3.8 %) contained green dyes. Cobalt Phthalocyanine was the most common dye, present at 66 fibers, followed by Hostasol Green G-K with three occurrences, while Irgazin Blue (Phthalocyanine), Indanthren Dark Blue, and Copper Phthalocyanine occurred in two fibers each. Sepisol Fast Blue occurred at a single fiber (Fig. 3). For a single fiber, two dyes were simultaneously identified: Hostasol Green G-K and Irgazin Blue (Fig. 4). The detected additives are widely used in industrial applications; all of them are dyes for plastics, nylon, cotton, and other materials. Five are known to be toxic for the biota, at least at a low toxicity level (Table 1).

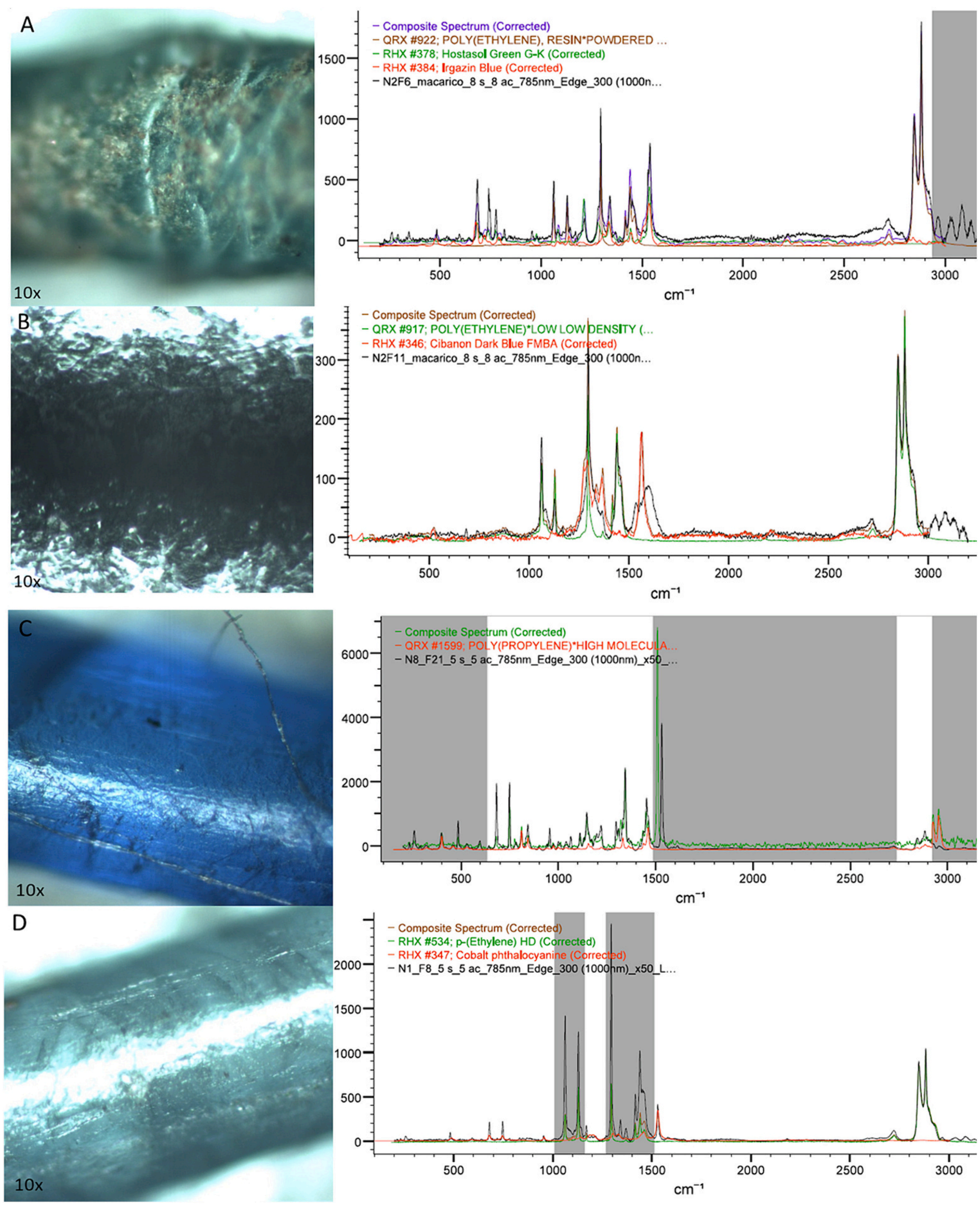

Fig. 4. Examples of color, texture, RAMAN spectra and associated additives from fibers removed from nests of the crested oropendola (*Psarocolius decumanus*). A: the single polyethylene containing two different dyes: Polychloro copper phthalocyanine (Hostasol green G-K) and Phthalocyanine (Irgazin blue); B: a low density polyethylene containing Dinaphtho(1,2,3-cd:3′,2′,1′-lm)perylene-5,10-dione (Cibanon dark blue); C: the single registered polypropylene fiber; D: a high density polyethylene containg Cobalt Phthalocyanine. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

Table 1
Name, synonyms, molecular formula, uses, toxicity and bibliographical references for the six dyes found in plastic fibers analyzed from nests of the crested oropendola (*Psarocolius decumanus*) in the Brazilian Amazon coast.

Name and synonyms	Formula	Uses	Toxicity/hazard	References
Cobalt Phthalocyanine (Phthalocyanine cobalt (II), Pigment Blue 74)	$C_{32}H_{16}N_8Co$	Paint industry, plastics, printing inks, rotogravure printing inks, coating materials	Hazardous by the 2012 OSHA Hazard Communication Standard. Respiratory Sensitization, Germ Cell Mutagenicity, Carcinogenicity	PubChem (2024b); Thermofisher (2024a); Ossila (2022)
Phthalocyanine (Irgazin Blue, Pigment Blue 16, Heliogen Blue 7560; Lionol Blue KW)	$C_{32}H_{18}N_8$	Industrial applications, especially in the coloring of plastics, paints, and coatings	Non-toxic or unavailable, not hazardous	PubChem (2024d); Thermofisher (2024b)
Dinaphtho(1,2,3-cd:3′,2′,1′-lm) perylene-5,10-dione (Indanthren Dark Blue, Indanthrone, Pigment Blue 65, Cibanon Dark Blue, Violanthrone)	$C_{34}H_{16}O_2$	Textile industry for coloring cotton, wool, and synthetic fibers	Low acute oral toxicity and low subchronic toxicity, moderate inhalation toxicity with potential lethality at high concentrations, copper toxicity in tissues, low risk of skin and eye irritation to rodents, harmful to aquatic life with long-lasting effects	PubChem (2024); European Chemical Agency (ECHA) (2024a)
Polychloro copper phthalocyanine (Phthalocyanine green G, Hostasol Green G-K, Acid Green 25, Phthalo Green, Pigment Green 7)	$C_{32}Cl_{16}CuN_8$	Textile industry for coloring protein fibers, such as wool, silk, and nylon	Low acute and chronic oral toxicity, mild skin and eye irritation, to rodents; low toxicity to microcrustaceans (*Daphnia magna*)	PubChem (2024c); European Chemical Agency (ECHA) (2024b); Sigma-Aldrich (2023); Information on Hazardous Chemicals and Occupational Diseases (Haz-Map) (2024a)
Sepisol Fast Blue	$C_{192}H_{248}N_{12}O_{14}S_4$	Textile, cosmetic, and plastic industries, among others. Can be used to dye cotton, polyester, nylon, and other textile fibers	Classified in category 4 for acute toxicity; toxicity to microcrustaceans (*Daphnia magna*)	PubChem (2024); Netherlands (2024); European Chemical Agency (ECHA) (2019); European Chemical Agency (ECHA) (2024b)
Copper Phthalocyanine (Hostaperm Blue, Neozapon Blue, Pigment Blue 15)	$C_{32}H_{16}CuN_8$	Textiles industries, plastics, paints, paper, leather, textile dyes, and printing	Low oral toxicity to rodents, low toxicity to microcrustaceans; suspected of carcinogenicity	PubChem (2024a); Information on Hazardous Chemicals and Occupational Diseases (Haz-Map) (2024b); Kodak (2021); International Labor Organization (ILO) and World Health Organization (WHO) (2021)

This study provides evidence of the widespread usage of polyethylene fibers by the crested oropendolas (*P. decumanus*) for nest construction along the Brazilian Amazon coast. The likelihood of plastic being used in nest construction depends not only on the abundance of debris near the nest but also on the availability of natural nesting material (Lavers et al., 2013; Witteveen et al., 2017). Consequently, the prevalence of plastics across all three sites (mangrove, grassy clearing, fishermen village) suggests this behavior may be increasingly common and linked to nylon fiber availability in the surrounding environment.

The dominance of blue fibers across all locations suggests a common source, likely linked to discarded fishing gear, already reported for the study site (Martinelli Filho and Monteiro, 2019; Silva, 2022). This aligns with the observation of accumulated fishing nets in the mangroves where all nests contained plastic fibers (Fig. 2), and all except one nest from the fishermen's village contained the same fibers. The lower proportion of plastics in the nests from the grassy clearing also supports the idea of a lower occurrence of plastic fibers in the environment since the site was dominated by other types of litter and natural fibers. Such correlation between the type and abundance of the surrounding plastic litter and its occurrence in the nests of marine and freshwater birds like *Morus bassanus*, *Ciconia ciconia*, *Larus dominicanus* and *Sula leucogaster* is already well documented (Lavers et al., 2013; Witteveen et al., 2017; Jagiello et al., 2018; O'Hanlon et al., 2019).

Seabirds choose nesting materials primarily based on shape and color (Tavares et al., 2019). Studies have already shown that fishing lines and ropes were also among the most prevalent items in brown booby nests (Lavers et al., 2013; Tavares et al., 2016). Our analysis confirmed that polyethylene was the primary polymeric component (97.5 %) of the analyzed fibers (Fig. 3). The dominance of high-density polyethylene suggests potential rigidity and durability of these fibers, possibly making them more suitable for nest construction, and an active selection and collection of such fibers by the birds. The choice for blue polyethylene fibers may be determined by the quality of material for nest construction, instead of the color, a hypothesis that demands future tests.

The Brazilian coast is widely impacted by plastic debris (Pegado et al., 2024), and some of these display potential hazards due to diverse associated contaminants (Cesar-Ribeiro et al., 2017). Moreover, the study site is known to bear moderate quantities of microplastics and fibers, mainly blue ones (Martinelli Filho and Monteiro, 2019). Adsorbed contaminants in microplastics were already reported for the Amazon coast, near the study area. Despite the relatively low concentrations, PAHs like naphthalene, phenanthrene, chrysene, pyrene, and fluoranthene are already adsorbed by microplastics in the region (Branco et al., 2023), which poses a potential risk of contamination by ingestion.

In a similar way, the fibers identified here also contain contaminants that are added during the industrial production of the colored nylon nets. Of the six identified dyes, five are known for at least a low toxicity degree in animal models, like rodents and microcrustaceans (*Daphnia magna*), but they have unknown effects on birds. While the range of toxicological effects on birds requires further investigation, previous studies have indicated potential health risks in animals, associated with some of these identified dyes (see Table 1).

The identification of Cobalt Phthalocyanine in a significant portion of the fibers (94.9 %) raises concerns about potential contaminant exposure for *P. decumanus*. This dye is listed as hazardous by the 2012 United States Occupational, Safety and Health Administration (OSHA) Hazard Communication Standard. Potential health effects include respiratory sensitization, germ cell mutagenicity, and carcinogenicity. Other identified dyes, such as Indanthren Dark Blue and Copper Phthalocyanine, have also been linked to slight toxicity in rodents and aquatic life (see Table 1).

The potential transfer of contaminants from plastic fibers to the crested oropendola eggs and chicks could be a significant concern. Studies have documented the presence of microplastics and associated contaminants in seabird eggs (van der Schyff et al., 2021), highlighting potential pathways for contaminant exposure in birds that utilize plastic as nesting materials.

The presence of tangled fibers and pieces of ropes could also lead to accidental entanglement of chicks or adults (Townsend and Barker, 2014), and such larger debris are also used by the crested oropendola. The ecological implications of incorporating plastics into nests require further study. While potential benefits like increased structural stability may be speculated, the increase in predation risk due to higher nest visibility was already observed (Corrales-Moya et al., 2023; Møller, 2017).

Our study is limited by the analysis of 36 abandoned, fallen nests, which difficult the elaboration of hypothesis on the effects of the high plastic load on both potential predators and *Psarocolius decumanus* itself. However, the effects of plastic additives on bird's health, as well as the effects on egg's viability are yet to be investigated since it may pose an important risk to these birds during different stages of their life cycle. At last, the presence, types and abundance of plastics in the nests of the crested oropendola may have a future potential as an indicator of plastic pollution in the surrounding environment (Tavares et al., 2016), an application which demands future research.

Funding

ACL was funded by the Coordenação de Aperfeiçoamento do Pessoal de Nível Superior (CAPES scholarship #88887.687937/2022-00). JEMF is supported by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq grant no. 408962/2023-02). A.T.S.F. and N.U.W. are supported by the Fundação de Amparo à Pesquisa do estado de São Paulo (FAPESP grant #2020/12050-6 and #2021/-4334-7, respectively). T.G. and N.U.W. are CNPq research fellows (# 308528/2022-00 and # 3085262021-0 respectively).

CRediT authorship contribution statement

Adrielle Caroline Lopes: Writing – review & editing, Writing – original draft, Visualization, Resources, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Maria Kuznetsova:** Writing – review & editing, Visualization, Validation, Supervision, Methodology, Investigation, Formal analysis, Data curation. **Anderson Targino S. Ferreira:** Writing – review & editing, Methodology, Investigation, Formal analysis, Data curation. **Niklaus Ursus Wetter:** Writing – review & editing, Visualization, Validation, Supervision, Resources. **Tommaso Giarrizzo:** Writing – review & editing, Visualization, Supervision, Investigation, Formal analysis, Conceptualization. **José Eduardo Martinelli Filho:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:

- The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgements

The authors thank the Biological Oceanography Laboratory staff for the aid during diverse steps of this research, especially for M.Sc. Leonardo M. S. Morais, M. Sc. Alysson A. L. Costa, M.Sc. Felipe Souza, M.Sc. Marcos F. B. C. Pereira, Thiago Maiandeua and Juan Damasceno. We acknowledge the aid of the anonymous reviewers and associate editor, which improved the overall quality of the accepted version of the manuscript.

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