



White Storks (*Ciconia ciconia*) as Indicators of Environmental Antimicrobial Resistance: A Review of Microbiological Insights

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Abstract: Wild birds, due to their ability to migrate over long distances and exploit highly anthropized habitats, act as ecological vectors in the circulation of pathogenic and antimicrobial-resistant bacteria, thereby influencing both human and animal health. The white stork (*Ciconia ciconia*), an opportunistic migratory species closely associated with human-altered habitats, plays a significant role in the dissemination of antimicrobial-resistant and potentially zoonotic bacterial strains across ecological and geographical boundaries. This review synthesizes microbiological evidence from recent studies evaluating bacterial isolation, resistance profiles, and genetic determinants in *C. ciconia* populations across Europe and neighboring regions. Reported findings indicate consistently high isolation rates of *Escherichia coli*, along with frequent detection of *Staphylococcus aureus*, coagulase-negative *Staphylococcus* spp., *Salmonella* spp., and other enteric bacteria. Genes such as *blaCTX-M*, *mcr-1*, *blaSHV-12*, *mecC*, and *tet* variants were commonly identified, reflecting complex multidrug-resistance mechanisms of clinical and ecological significance. Foraging at solid waste disposal sites and proximity to human settlements were consistently associated with increased resistance prevalence. Ecological and behavioral factors, including parental feeding habits and habitat type, were shown to modulate both microbiota composition and the resistome structure. Collectively, these findings emphasize the role of the white stork as a bioindicator of environmental antimicrobial resistance and a potential ecological link in the transmission of zoonotic and resistant bacteria. The inclusion of wild avifauna in AMR surveillance strategies remains essential for understanding the complex interactions among humans, domestic animals, and the environment under the One Health paradigm.

Introduction

This review synthesizes microbiological evidence from recent studies published between 2015 and 2025 that investigated bacterial diversity, isolation rates, and antimicrobial resistance in white storks (*Ciconia ciconia*). These studies encompass both free-ranging populations and individuals admitted to Wildlife Rehabilitation Centres (WRCs), providing an integrative view of how environmental exposure, captivity, and anthropogenic factors influence the occurrence and dissemination of resistant or potentially zoonotic bacteria.

Particular attention is given to commensal and opportunistic pathogenic species, including *Escherichia coli*, *Salmonella* spp., *Pseudomonas* spp., and *Staphylococcus* spp., which are frequently reported in avian hosts and are of growing concern in both veterinary and public health contexts. This review compares findings on bacterial prevalence and antimicrobial susceptibility patterns to identify recurring resistance mechanisms and to assess their ecological and clinical significance. Particular focus is given to multidrug-resistant (MDR) and extended-spectrum β -lactamase (ESBL)-producing isolates, highlighting their association with factors such as feeding behavior, habitat type, and proximity to anthropogenic waste sources.

Furthermore, the review explores the broader role of Wildlife Rehabilitation Centres as essential epidemiological surveillance nodes within the One Health framework. By serving as convergence points for birds originating from diverse geographic regions and often subjected to physiological stress, these facilities offer unique opportunities for the early detection of emerging pathogens and the monitoring of antimicrobial resistance circulation across wildlife populations. Collectively, this synthesis aims to enhance understanding of the ecological drivers shaping the resistome of *Ciconia ciconia* and to underline its relevance as a sentinel species for environmental health assessment.

Materials and methods

A comprehensive literature search was conducted for studies published between January 2015 and November 2025 using the databases PubMed, Scopus, and Web of Science. The following combinations of keywords and Boolean operators were applied: ("white stork" OR "*Ciconia ciconia*") AND ("microbiota" OR "bacteria" OR "microbiological analysis" OR "antimicrobial resistance" OR "antibiotic resistance" OR "pathogenic bacteria" OR "wildlife sentinel").

Additional references were identified through manual searches of bibliographies from relevant papers.

Results and discussions

All twenty reviewed studies were observational, reflecting early research on *Ciconia ciconia* as a sentinel and potential vector for zoonotic bacteria and environmental antimicrobial resistance (AMR), highlighting its key role in pathogen transmission at the wildlife-livestock-human interface. Several studies employed comparative or cross-sectional approaches to assess microbiological and molecular profiles across diverse environmental contexts, including landfills, agricultural zones, and natural habitats. Representative examples include [Abdullahi et al., 2023a–c; Martín-Maldonado et al., 2022; Migura-García et al., 2020; Camacho et al., 2016], which explicitly investigated how varying degrees of anthropogenic influence shape bacterial diversity and antimicrobial resistance patterns. In contrast, other studies such as [Ivan et al., 2025] and [Szczepeńska et al., 2015] adopted more descriptive, site-specific designs, focusing on the characterization of culturable isolates within localized or rehabilitation settings and thereby providing valuable complements to the broader ecological assessments.

Geographically, most research originated from Southern and Central Europe, with Spain emerging as the leading contributor. Fourteen of the twenty studies were conducted across Spanish regions, evidencing a sustained research focus on Iberian stork populations. Additional reports from Poland, Portugal, Romania, Algeria, and Türkiye [Szczepeńska et al., 2015; Santos et al., 2025; Ivan et al., 2025; Loucif et al., 2022; Uslu et al., 2024] broaden the understanding of this sentinel species' distribution and its interaction with varied ecological and anthropogenic pressures throughout Europe and North Africa. Building upon the previously mentioned methodological and geographical foundations, the reviewed literature collectively reveals consistent patterns linking habitat type, feeding ecology, and antimicrobial resistance dissemination in *Ciconia ciconia*. Across twenty observational studies published between 2015 and 2025, more than 3,000 individual samples were analyzed from diverse ecological contexts throughout Europe and North Africa. Most investigations were conducted in Spain ($n = 14$), followed by Poland, Portugal, Romania, Türkiye, and Algeria, reflecting both the species wide distribution and its central relevance within integrated One Health surveillance frameworks.

The earliest study considered in this review, conducted by [Szczepeńska et al., 2015] in Poland, revealed that even nestlings from colonies located near polluted areas harbored *C. jejuni* and *C. coli* strains, showing notable resistance to fluoroquinolones and tetracycline. Although the overall prevalence (5.3% and 2.3%) was modest, the clear environmental gradient, higher infection rates in anthropogenically influenced colonies, was a critical early indication that human-related contamination directly shapes the microbial ecology of wild birds. Importantly, the detection of key virulence genes (*fliA*, *cadF*, *cdtB*, *iam*) revealed that these isolates were not merely environmental contaminants but potentially zoonotic strains capable of persistence and transmission.

Building on earlier findings, [Camacho et al., 2016] compared natural and landfill colonies, reporting high *E. coli* prevalence ($\approx 90\%$) and multidrug resistance (35–58%) in both settings, with landfill isolates showing greater resistance to β -lactams and fluoroquinolones. The concurrent detection of *Salmonella* spp. highlighted exposure to human-derived pathogens in waste-fed populations, positioning landfills as key hotspots for resistant enteric bacteria.

A major advancement came from [Höfle et al., 2020], who analyzed 467 cloacal swabs from Spanish storks and found cephalosporin-resistant *E. coli* in 8.8% of samples. All isolates were resistant to ampicillin, cefotaxime, and nalidixic acid, and over half (54%) were multidrug resistant. Detected genes included *blaCTX-M-1* (43.5%), *blaCMY-2* (19.6%), *blaSHV-12* (15.5%), and *blaCTX-M-32* (5.6%), with one isolate simultaneously carrying *mcr-1*, *blaCMY-2*, and *blaCTX-M-32*. Plasmid replicons (*IncI*, *IncFIB*, *IncW*) indicated strong potential for horizontal gene transfer from anthropogenic sources such as livestock waste and wastewater, confirming storks as vectors of clinically relevant AMR genes.

Simultaneously, [Martín-Maldonado et al., 2020] reported *Salmonella* spp. from landfill-foraging storks with high ciprofloxacin, nalidixic acid, and colistin resistance, an alarming profile of last-resort antimicrobial exposure, with nearly 87% of isolates being multidrug resistant. [Migura-García et al., 2020] further demonstrated molecular overlap of AMR genes between storks and livestock, identifying identical *ST10 E. coli* clones and plasmids (*IncX4*, *IncHI2*, *IncI2*) carrying *mcr-1* and *blaCTX-M*, evidencing cross-species gene flow. The metagenomic work of [Jarma et al., 2021] and [Pineda-Pampliega et al., 2021] expanded this ecological perspective, confirming that landfill exposure enriches the resistome (*int11*, *sul1*, *blaTEM*, *tetW*, *qnrS*) and promotes co-selection of virulence and resistance traits through APEC-like plasmids (*iroN*, *ompT*, *blyE*, *iutA*, *iss*).

[Loucif et al., 2022] provided the first evidence of carbapenemase- and colistin-producing Enterobacterales in North African storks, including *blaOXA-48*, *blaNDM-5*, and *mcr-1*, showing that AMR dissemination in migratory birds now spans continental scales. Geographically, the range and intensity of AMR detection have grown markedly. At the same time, studies from captive populations [Martín-Maldonado et al., 2022; Ivan et al., 2025] revealed that even controlled environments sustain low but persistent resistance, likely through commensal exposure. The Romanian study by [Ivan et al., 2025] offered the first microbiological characterization of *C. ciconia* in that region, isolating 52 bacterial strains, mainly *E. coli* (95.5%) and *Salmonella enterica* (18.2%), alongside *Kocuria varians*, *Serratia fonticola*, *Staphylococcus sciuri*, and *Pseudomonas aeruginosa*. Although resistance genes were not assessed, the presence of multiple zoonotic and opportunistic species underscored the value of wildlife rehabilitation centers as AMR surveillance sites.

The series of studies by [Abdullahi et al., 2023a–c] broadened the taxonomic focus, showing that Gram-positive bacteria also mirror landfill effects. *Staphylococcus aureus* and coagulase-negative staphylococci displayed higher resistance to penicillin and tetracycline and carried *blaZ*, *mecA*, *tetK*, and *tetM*, including the first *mecC*-positive *S. lentus* in wild birds. Subsequent molecular analyses by [Márquez-Álvarez et al., 2023] and [Sacristán-Soriano et al., 2024] identified mobile genetic elements (*Tn21*, *IS26*, *int11/int12*) associated with *blaCTX-M* and *tetM*, linking landfill exposure to active gene mobilization.

Later studies in Türkiye and Spain [Uslu et al., 2024; Cardona-Cabrera et al., 2025a,b] confirmed high AMR prevalence associated with landfill feeding, detecting mercury-resistance transposons and even caffeine in plasma, clear markers of anthropogenic waste ingestion. Conversely, [Łopińska et al., 2025] emphasized that waterborne exposure may be equally important in Central Europe, while [Santos et al., 2025] described a milder scenario in Portugal, with moderate resistance (36% ampicillin, 12% tetracycline) and low MDR (5.3%) in rehabilitated birds. Collectively, these findings highlight geographic variability in AMR dynamics, influenced by local environmental pressures and pollution pathways.

Conclusions

Collectively, the reviewed evidence demonstrates that the white stork (*Ciconia ciconia*) serves as a valuable bioindicator of environmental antimicrobial resistance within the One Health framework. Across Europe and North Africa, studies consistently reveal a strong link between habitat type, foraging behavior, and the prevalence of resistant bacteria and resistance genes. Landfill-associated populations repeatedly exhibit higher rates of multidrug-resistant and ESBL-producing *Escherichia coli* and *Salmonella* spp., alongside mobile genetic elements such as *mcr* and *blaCTX-M*, underscoring the role of anthropogenic waste as a major reservoir and amplifier of AMR. At the same time, findings from natural colonies and rehabilitation centers highlight that resistant and potentially zoonotic bacteria persist even in less contaminated environments, reflecting the pervasive reach of antimicrobial selection pressures. Altogether, white storks integrate ecological, microbiological, and molecular signals of environmental contamination, making them an invaluable sentinel species for tracking the dissemination of resistance determinants across ecosystem boundaries. Future research combining genomics, spatial ecology, and standardized surveillance protocols will be crucial to fully elucidate their role in AMR transmission dynamics and to inform more effective environmental health monitoring strategies.