Science of the Total Environment 985 (2025) 179742

Contents lists available at ScienceDirect

Science of the Total Environment

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

Global distribution of antimicrobial resistance genes in *Escherichia coli* isolated from wild animals using genomes available in public databases

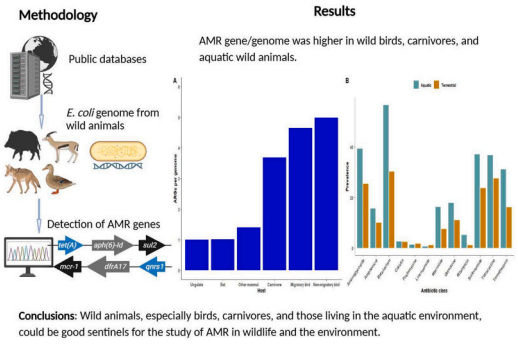
Andnet Yirga Assefa [a], Biel Garcias [b,1], Evangelos Mourkas [c], Rafael A. Molina-López [d], Laila Darwich [b,*]

[a] Department of Veterinary Epidemiology and Public Health, School of Veterinary Medicine, Bahir Dar University, Bahir Dar, Ethiopia
[b] Departament Sanitat i Anatomia Animals, Veterinary Faculty, Universitat Autònoma de Barcelona (UAB), Cerdanyola del Vallès, Spain
[c] Zoonosis Science Center, Department of Medical Sciences, Uppsala University, Uppsala, Sweden
[d] Wildlife Rehabilitation Center of Torreferrusa, Barcelona, Spain

HIGHLIGHTS

- *E. coli* isolates from low- and middle-income regions harbored higher number ARGs.
- Wild birds and carnivores harbored higher ARGs than other wild animals.
- The ARGs were associated with resistance to common and last-resort antibiotics
- The number of ARGs per genome was higher in aquatic wild animals than terrestrial ones.
- Wild birds, carnivores and aquatic animals could be AMR sentinels in the environment.

GRAPHICAL ABSTRACT



ARTICLE INFO

Editor: Ewa Korzeniewska

Keywords:
Antimicrobial resistance
Antimicrobial resistant genes
Escherichia coli
Environment
Genomes
Wild animals

ABSTRACT

Antimicrobial resistance (AMR) is a pressing worldwide health challenge fueled by the improper and/or excessive use of antimicrobials in humans and animals. Wild animals can acquire AMR from waste released into the environment, being a good bioindicator of AMR pollution in that compartment. This study aimed to estimate the global distribution of antimicrobial-resistant genes (ARGs) in *Escherichia coli* (*E. coli*) isolated from wild animals (birds and mammals) and to assess their role as sentinels for the study of AMR in the environment. A total of 4436 *E. coli* genomes were retrieved from three public databases and screened for ARGs using ResFinder, revealing 159 unique ARGs. The tetracycline-resistant gene *tet(A)* was the most frequently detected (22.1 %). The highest burden of ARGs per genome was identified in Asia (5.9), followed by Africa (5.2) and South and Central America (5). Wild birds and carnivores harbored more ARGs per isolate than wild ungulates, bats and other mammals. Additionally, wild animals inhabiting aquatic environment carried a higher number ARGs per genome compared to terrestrial species. Furthermore, genomes from those groups of animals showed a higher level of predicted resistance to classes of antibiotics widely used in humans and animals, including beta-lactams,

* Corresponding author.
E-mail addresses: biel.garcias.puigserver@gu.se (B. Garcias), evangelos.mourkas@uu.se (E. Mourkas), rafael.molina@gencat.cat (R.A. Molina-López), laila.darwich@uab.cat (L. Darwich).
[1] Current address: Department of Microbiology and Immunology, University of Gothenburg, Gothenburg, Sweden.

https://doi.org/10.1016/j.scitotenv.2025.179742
Received 25 March 2025; Received in revised form 22 May 2025; Accepted 22 May 2025
Available online 27 May 2025

aminoglycosides, tetracyclines, sulfonamides, trimethoprim, and last-resort antibiotics like colistin and carbapenems. Given the higher burden and their presence in different ecosystems, wild animals, especially birds, carnivores, and those living in the aquatic environment, could be good sentinels for the study of AMR pollution in the environment.

1. Introduction

Antimicrobial resistance (AMR) is one of the major global public health and development threats. Only in 2019, AMR contributed to 4.95 million deaths globally, with bacterial AMR alone accounting for 1.27 million deaths (Murray et al., 2022). This number is expected to rise to 10 million by 2050 with the highest mortality will be in low and middle-income countries (LMICs) (O'Neil, 2014). Furthermore, AMR poses a significant economic burden by increasing healthcare costs due to the need for expensive, last-resort drugs and prolonged hospital stays. It also impacts animal health, adding costs to the agricultural sector. The World Bank estimates that by 2050, AMR could result in an additional US$ 1.2 trillion in healthcare costs and reduce global livestock production by 3–8 % annually, potentially reaching 11 % in a high-impact scenario (World Bank, 2017).

Understanding the dynamics of AMR demands multisectoral surveillance involving humans, animals, and the environment, including wildlife. However, the current surveillance systems largely focused on only livestock and humans (Anjum et al., 2021). Although a low level of AMR is expected in wild animals due to their rare exposure to antimicrobials, the overlap between habitats facilitates the transmission of resistant bacteria and genes across different habitats (Dias et al., 2015; Jones et al., 2008). Wild animals could easily acquire the AMR bacteria and genes released from humans, domestic animals, aquaculture and plant agriculture, where antimicrobials are widely used (Atterby et al., 2017; Schar et al., 2020; Miller et al., 2022), and can serve as sentinels, reservoirs and spreaders of AMR bacteria and genes to humans and other animals (Allen et al., 2010; Wu et al., 2018).

Wild animals, especially those living at the interface of natural and human-modified environments, could acquire AMR bacteria and ARGs through contaminated water, soil, or direct contact with domestic animals and humans (Guenther et al., 2011). Such exposure is prominent in LMICs, and exacerbated by limited access to health care and veterinary services, poor waste management, extensive use of antimicrobials in agriculture, and close interaction between humans and animals (Khalid et al., 2018; Sulis et al., 2022; Van Boeckel et al., 2015). Therefore, the burden of AMR in wild animals is expected to be higher in those regions than in high-income countries (HICs), where antimicrobial use and waste management are better regulated, and there is better access to health care and veterinary services (Olaru et al., 2023; Khalid et al., 2018; Van Boeckel et al., 2015).

Antimicrobial resistance has been detected in various wild mammals and birds, with the prevalence and patterns of resistance varying across species and locations (Oravcova et al., 2013; Bonnedahl and Järhult, 2014; Carroll et al., 2015). However, only a few studies include both species and compare the prevalence of AMR among them. Some studies have found a higher prevalence of AMR in wild birds than in wild mammals (Łopucki et al., 2024; Smith et al., 2014), while other studies reported a higher burden in wild mammals (Swift et al., 2019; Darwich et al., 2019).

Habitats may also influence wildlife species' exposure to AMR and the extent of its dispersion in the environment (Vittecoq et al., 2016; Swift et al., 2019). Aquatic habitat plays a significant role in the dispersal of antibiotics and AMR bacteria into natural ecosystems. For instance, studies on marine mammals have demonstrated that they carry a highly diverse range of AMR bacteria (Schaefer et al., 2009), and the prevalence of these bacteria is increasing alarmingly (Wallace et al., 2013).

Additionally, as natural wetlands are lost globally, water birds have become increasingly reliant on alternative and artificial habitats, such as wastewater treatment wetlands (Murray and Hamilton, 2010). This reliance may facilitate the transmission of human origin AMR bacteria to wild birds. Therefore, aquatic habitats may be more affected by AMR contamination than terrestrial environments. However, comparing the burden of AMR bacteria between aquatic and terrestrial wild animals is challenging since only a few studies include animals from both habitats (Vittecoq et al., 2016). Furthermore, recent studies have shown that the AMR profile of *E. coli* isolated from sewage samples is correlated with AMR in *E. coli* from the corresponding populations (Delgado-Blas et al., 2021; Huijbers et al., 2020; Hutinel et al., 2019). Thus, *E. coli* could be an important bioindicator to assess the impact of human activity on the environment, including wild animals (Anjum et al., 2021), and indicates that aquatic animals are at higher risk of acquiring AMR.

Studies involving genetic analysis of AMR in wildlife are limited, and they predominantly rely on culture techniques (Blanco-Peña et al., 2017; Jiang et al., 2013). For instance, phenotypic methods are the most widely used for systematic surveillance of AMR in developed countries (Aerts et al., 2019) and for epidemiological studies outlining resistance in low and middle-income countries (Van Boeckel et al., 2019). However, a recent decrease in price and an increase in the ease of metagenomics have allowed a wider use of these technologies, leading to more genomes available in public repositories. This increase in genomic data provides a wealth of information to track trends and prevalence of ARGs (Pires et al., 2021). Nevertheless, most isolates in public repositories are from humans, with less emphasis on animals (Pires et al., 2022), and even much less on wild animals. Although some studies have analyzed publicly available genomes to assess AMR in different bacteria across various hosts, no study has specifically focused on *E. coli* from wild animals.

Therefore, this study aims to estimate the global distribution of ARGs in *E. coli* isolated from wild animals using genomes available in public databases. It also seeks to determine the predicted prevalence of ARGs to different classes of antibiotics among continents, host groups, and habitats of wild animals, and to assess the role of wild animals as sentinels for studying AMR pressure in the environment.

2. Material and methods

2.1. Downloading genome assemblies and metadata

Genomic assemblies deposited until 31 December 2023 were collected from three public databases: Enterobase, the Nucleotide Database of the National Center for Biotechnology Information (NCBI), and the Pathosystems Resource Integration Center (PATRIC). Genomes from NCBI and Enterobase were downloaded directly from the respective websites, while the genomes from PATRIC were downloaded through the PATRIC command line interface. Accession numbers and Biosamples were used to identify genomes deposited in more than one database. Repeated genomes were removed from the dataset. The associated metadata were downloaded from (i) the Enterobase search strain website; (ii) the NCBI pathogen detection website; and (iii) the PATRIC taxon view website.

2.2. Standardization of the metadata

The metadata from the three databases was inspected manually, and entries that did not report the origin of isolates (country) were removed. The entry was further curated using keywords including source niche,

host name, and source type to exclude entries that did not meet the criteria that the isolates were from wild animals. Only genomes from wild birds and mammals were included in this study, as most of the genomes from public repositories were from species belonging to these taxonomic groups and were proportionally represented. Genomes from amphibians were excluded since only 46 genomes were retrieved from the three databases. Besides, isolates from fish were not considered as there was no clear information in the metadata whether the fish were from aquaculture or capture fishery, which may cause misrepresentation of the wildlife.

Other columns were added manually to the metadata. Wild animals were grouped as birds and mammals and further classified as migratory birds, non-migratory birds, ungulates, carnivores, bats, and other mammals. The classification of birds as migratory and non-migratory was based on (Del Hoyo, 2020; Lincoln et al., 1998; Spina et al., 2023; UNEP-WCMC, 2024), and partially migratory birds were considered as migratory in the classification. The host group "other mammals" includes all wild mammals that could not be assigned to ungulates, carnivores, or bats. Based on the habitat in which they live, wild animals were categorized as aquatic and terrestrial animals, and according to the country of origin of the *E. coli* isolates a new variable "continent" was added. List of bird and mammal species included in the study, along with their respective host categories and natural habitats is reported in the supplementary table (Table S1).

The final metadata table contains the following information: the assembly ID (name of the assembly ID as identified in the database), name of the database where the genomes were retrieved, host (whether the wild animal was bird or mammal), host group (classified as ungulates, carnivores, bats, other mammals, migratory birds and non-migratory birds), habitat of the wild animals (terrestrial versus aquatic environment), year of strain isolation, country and continent of isolations, type of ARGs, and a class of antibiotics for which the ARGs are predicted to be resistant.

2.3. Detection of ARGs

ABRicate (version 1.0.1), using the ResFinder database, was used to screen ARGs found in the genomes (Bortolaia et al., 2020). Matches with an identity and coverage of >80 % were considered a positive hit, which is default threshold for ABRicate (Lipworth et al., 2024; Vieira et al., 2023).

2.4. Curation of predicted phenotypes

For each ARG, the predicted resistance to antibiotic classes was retrieved from the ResFinder database (https://bitbucket.org/genomicepidemiology/resfinder_db/src/master/notes.txt). Overall, predicted resistance to 12 classes of antibiotics (Aminoglycoside, Amphenicol, Beta-lactam, Fosfomycin, Lincosamide, Macrolide, Colistin, Rifampicin, Sulfonamide, Tetracycline, Trimethoprim, and Quinolone) was identified. A list of ARGs and their respective predicted resistance phenotypes is described in the supplementary table (Table S2).

2.5. Data analysis and statistics

R-software (version 4.3.2) was used to explore and analyze the dataset. R-packages *tidyr* and *dplyr* were used to organize the data matrix and *ggplot2* was used to generate graphs (R Core Team, 2024). Kruskal-Wallis test was used to assess whether the difference in the number of ARGs per genome between continents and host groups was statistically significant ($p < 0.05$), and pairwise Wilcoxon test was used as a post-hoc test to identify between which groups exactly the difference occurred. Besides, Wilcoxon test was used to assess whether the difference in the number of ARGs between habitats of wild animals was statistically significant. Logistic regression was used to assess whether the difference in the prevalence of specific ARGs and predicted resistance to antibiotic classes per continent, host groups, and habitats was statistically significant ($p < 0.05$).

3. Results

3.1. Global E. coli genome dataset from wildlife

Overall, 4436 unique genome assemblies were retrieved from the three public databases: the majority of the genomes 70.2 % were from Enterobase (n = 3114), followed by 28 % from NCBI (n = 1243), and only 1.78 % were from PATRIC (n = 79) (Fig. S1A). About 58.1 % of the genomes were recovered from birds, while the remaining 41.9 % were from mammals (Fig. S1B). According to the habitat of wild animals, 56 % of the retrieved genomes were from terrestrial animals (n = 2483) and the remaining 44 % were from aquatic animals (n = 1953) (Fig. S1C).

Geographically, the largest part of the isolates was from North America (n = 1571, 35.4 %) and Oceania (n = 1408, 31.7 %) (Fig. S2A), with most isolates from the United States of America (n = 1498, 33.8 %) and Australia (n = 1348, 30.4 %) (Fig. S2B). The dataset includes genomes isolated from 1970 to 2023. The highest number of genomes was from 2019 (n = 958, 21.6 %), followed by 2017 (n = 693, 15.6 %) and 2016 (n = 538, 12.1) (Fig. S2C). Based on the host group, the highest number of genomes was from non-migratory birds (n = 1321, 29.8 %) and migratory birds (n = 1258, 28.4 %), while the lowest was from bats (n = 179, 4 %) (Fig. S2D).

A total of 15,237 ARGs were identified, including 159 unique ARGs. However, *mdf(A)* gene was removed from the dataset since it is an intrinsic resistance gene of *E. coli*, and the remaining genes were considered for further analysis. The most abundant ARGs detected were *tet(A)* (22.1 %), *sul2* (21.2 %), *aph(6)-Id* (21.2 %), *aph(3″)-Ib* (20.6 %), $bla_{\text{TEM-1B}}$ (18.4 %), *sul1* (14.2 %), *mph(A)* (10.8 %), and *tet(B)* (9.7 %) (Fig. 1A). The overall data analysis showed that 41.8 % of the genomes harbored at least one ARG associated with beta-lactam resistance, followed by aminoglycoside and tetracycline (31.5 % each), sulfonamide (29.6 %), and trimethoprim (22.7 %). Only 0.9 % of the genomes contained ARGs linked to lincosamide resistance (Fig. 1B).

3.2. Geographic distribution of ARGs

There was a significant difference ($p < 0.05$) in the number of ARGs per genome among different continents, with the highest in Asia (5.9), followed by Africa (5.2) and South and Central America (5), and the lowest in North America (2.2) (Fig. 2A). Although the prevalence was different, the genomes from Asia, Europe and Oceania had at least one ARG associated with all the 12 classes of antibiotics. Africa and Asia had the highest number of ARGs associated with resistance to most classes of antibiotics, including beta-lactam, aminoglycoside, tetracycline, sulfonamide, trimethoprim and quinolone. Additionally, genomes from Asia showed the highest frequency of ARGs resistance to amphenicol, colistin and fosfomycin. Resistance per class of antibiotics was highest for beta-lactam across all continents, with 73.6 % in Africa, 69.2 % in Asia, 53.1 % in Oceania, 37.8 % in South and Central America, 37.4 % in Europe and 28.6 % in North America, and these differences were statistically significant. Notably, no ARGs were detected for rifampicin, colistin and lincosamide in Africa; for colistin in North America; and for lincosamide in South and Central America (Fig. 2B).

The most abundant ARGs identified slightly varied among continents. For instance, in Africa the most abundant ARGs detected were $bla_{\text{CTX-M-15}}$ (63.6 %); in Asia, *tet(A)* (42.4 %); in Europe, *tet(A)* (22.3 %); in North America, *aph(6)-Id* (15.8 %); in Oceania, $bla_{\text{TEM-1B}}$ (27.5 %), and in South and Central America, *tet(A)* (29.3 %) (Fig. 2C). Among ARGs associated with resistance to clinically important antibiotics, ESBL genes were the most widely distributed, with $bla_{\text{CTX-M}}$ being the most frequently detected. Europe and Oceania were the only continents where the two colistin-resistant genes (*mcr-1* and *mcr-9*) were detected. However, the prevalence of *mcr-1* was extremely high in Asia (26.3 %),

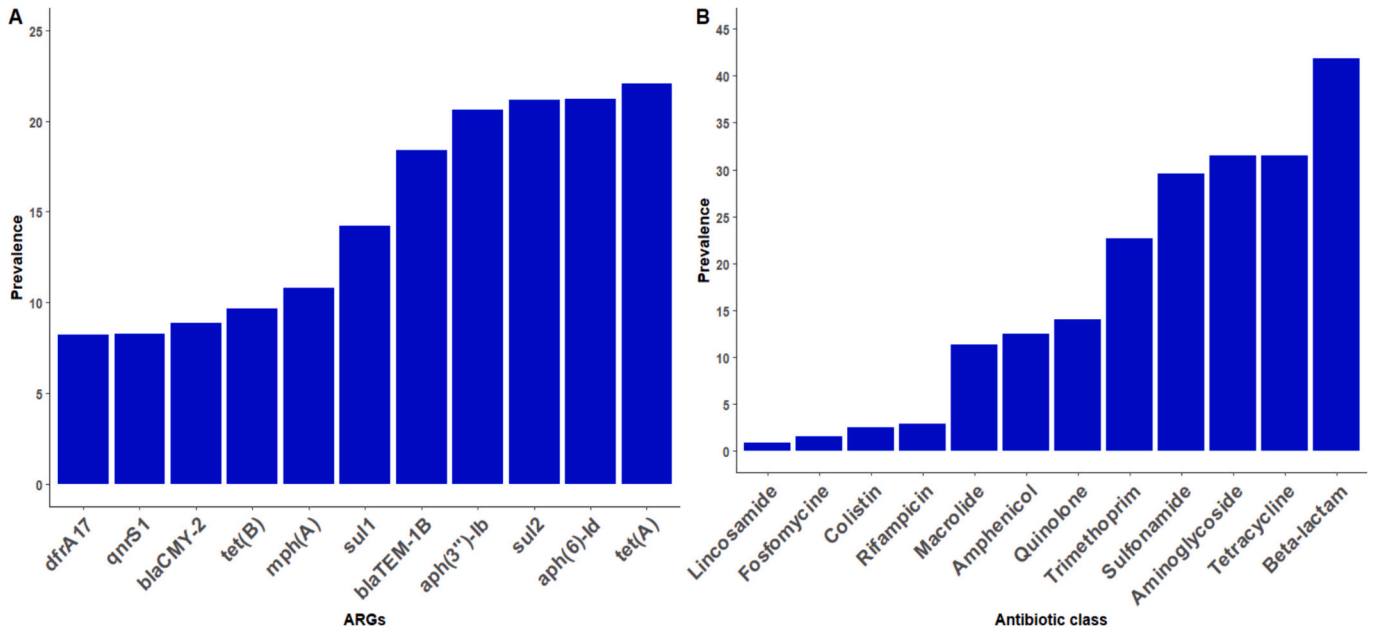


Fig. 1. A: Barplot showing the most abundant ARGs detected (prevalence ≥8 %). B: Barplot showing the predicted resistance per class of antibiotics.

Fig. 2. Geographical distribution of ARGs. A: Barplot showing the number of ARGs per isolate per continent. B: Heatmap showing the prevalence of resistance to antibiotic classes per continent (N. America: North America, S&C America: South and Central America). C: A bar plot showing the ARGs having the highest relative abundance per continents colored by antibiotic class (top 10 abundant ARGs).

while *mcr-9* was higher in South and Central America (12.2 %). However, this difference was not statistically significant ($p > 0.05$). Most carbapenem-resistant genes, including bla_{OXA-48}, $bla_{OXA-245}$, bla_{NDM-1}, bla_{NDM-5}, bla_{VIM-1}, bla_{VIM-4} and bla_{KPC-2}, were identified in Europe, but their prevalence was lower than 2 %. The highest prevalence of bla_{NDM-5} was observed in South and Central America (13.4 %) and Asia (5.1 %), whereas bla_{IMP-4} was detected only in Oceania, with a prevalence of around 6.8 % (Fig. S3).

3.3. Wild birds carry more ARGs than wild mammals

Wild birds harbored more ARGs per genome than mammals, with 4.8 and 1.5 ARGs per isolate, respectively. Among groups of wild animals, non-migratory birds had the highest ARGs per genome (5), followed by migratory birds (4.7) and carnivores (3.7), while the lowest was in ungulates and bats with around 1 ARG per genome (Fig. 3A).

Overall, wild birds (migratory and non-migratory) and carnivores harbored isolates with the highest level of resistance (>30 %) to the most common classes of antibiotics, including beta-lactams, tetracyclines, aminoglycosides and sulfonamides. In contrast, bats, ungulates and other mammals exhibited lower level of resistance (< 21 %) for all antimicrobials, with no resistance associated with rifampicin and lincosamides detected. Besides, no resistance to macrolides, colistin and fosfomycin was found in bats, and no resistance to colistin was observed in ungulates (Fig. 3B).

Differences in the distribution of ARGs among host groups were observed. The most frequent ARGs observed in carnivores, migratory and non-migratory birds were *tet(A)*, *sul2*, *aph(6)-Id* and bla_{TEM-1B}, with the latter also frequently detected in bats. Aminoglycoside-resistant genes (*aph(6)-Id* and *aph(3″)-Ib*) showed relatively similar within-host group prevalence across all host groups. However, their prevalence varied between host groups, with the highest frequencies observed in carnivores, migratory and non-migratory birds (Fig. 3C). Except for other mammals, ESBL genes were found in all host groups, with bla_{CTX-M} genes being the most frequently detected. Specifically, a high prevalence of $bla_{CTX-M-15}$ was observed in migratory birds (16.9 %). Both migratory and non-migratory birds were the only host groups that harbored *mcr-1* and *mcr-9* genes, with the prevalence being higher in non-migratory birds. In contrast, no colistin-resistant gene was detected in bats and ungulates. The carbapenem-resistant gene (bla_{IMP-4}) was abundant in non-migratory birds (7.3 %), whereas bla_{OXA-48} was more prevalent in migratory birds (1.6 %). Remarkably, no carbapenem-resistant gene was found in genomes from carnivores and other mammals (Fig. S4).

3.4. Habitat ecology drives the distribution of ARGs in wildlife

The distribution of ARGs also varies among the habitats where wild animals live. Wild animals living in aquatic habitats had 4.5 ARGs per genome, while their counterparts in the terrestrial environment had only 2.6 ARGs. With the exception of fosfomycine and lincosamide, resistance to all classes of antibiotics was higher in aquatic animals than in terrestrial ones. The highest level of resistance detected in both groups of

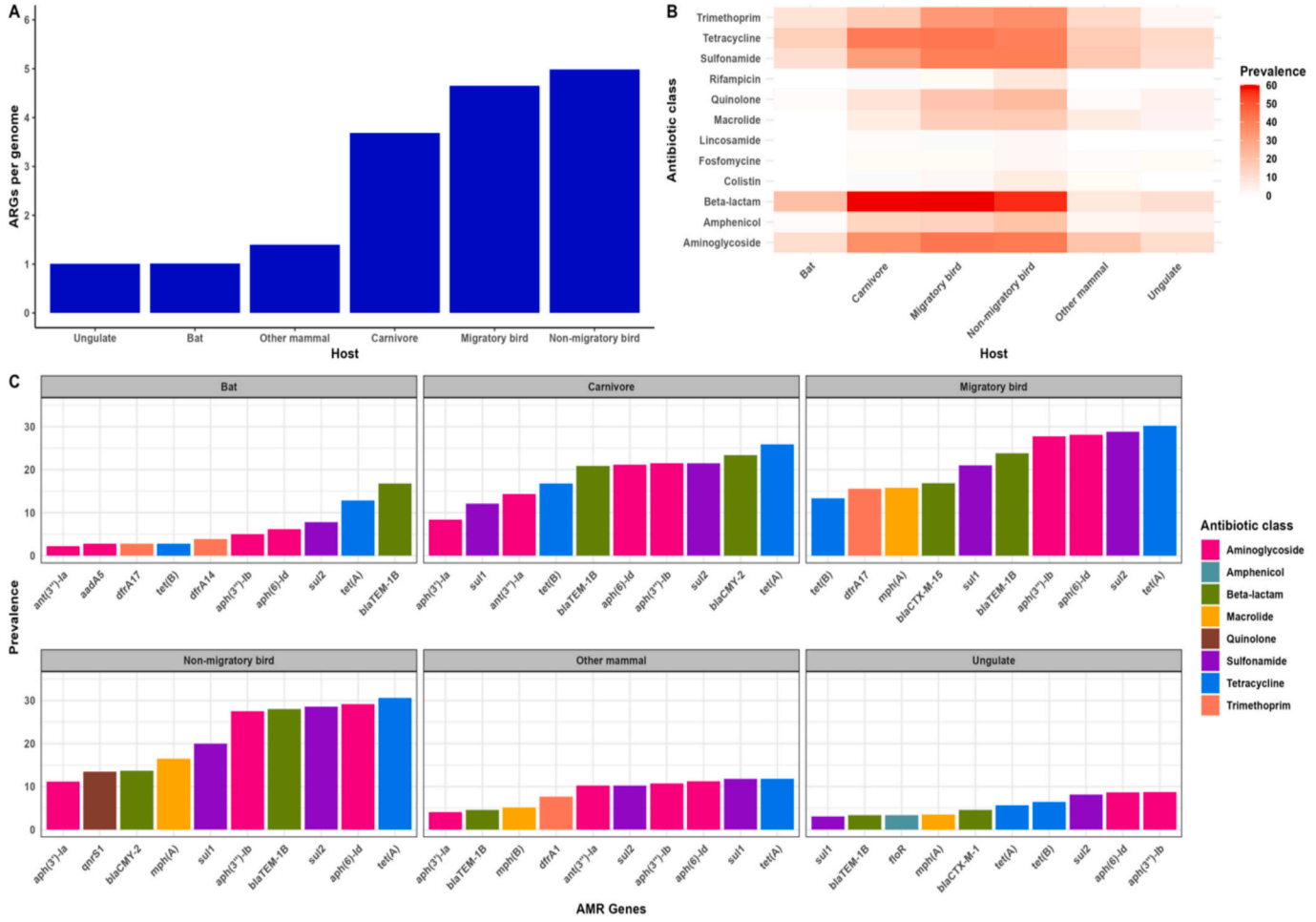


Fig. 3. Distribution of ARGs among host groups. A: Barplot showing the number of ARGs per isolate per host group. B: Heatmap showing the prevalence of resistance to antibiotic classes per host groups. C: Barplot showing the ARGs having the highest relative abundance per host groups colored by antibiotic class (top 10 abundant ARGs).

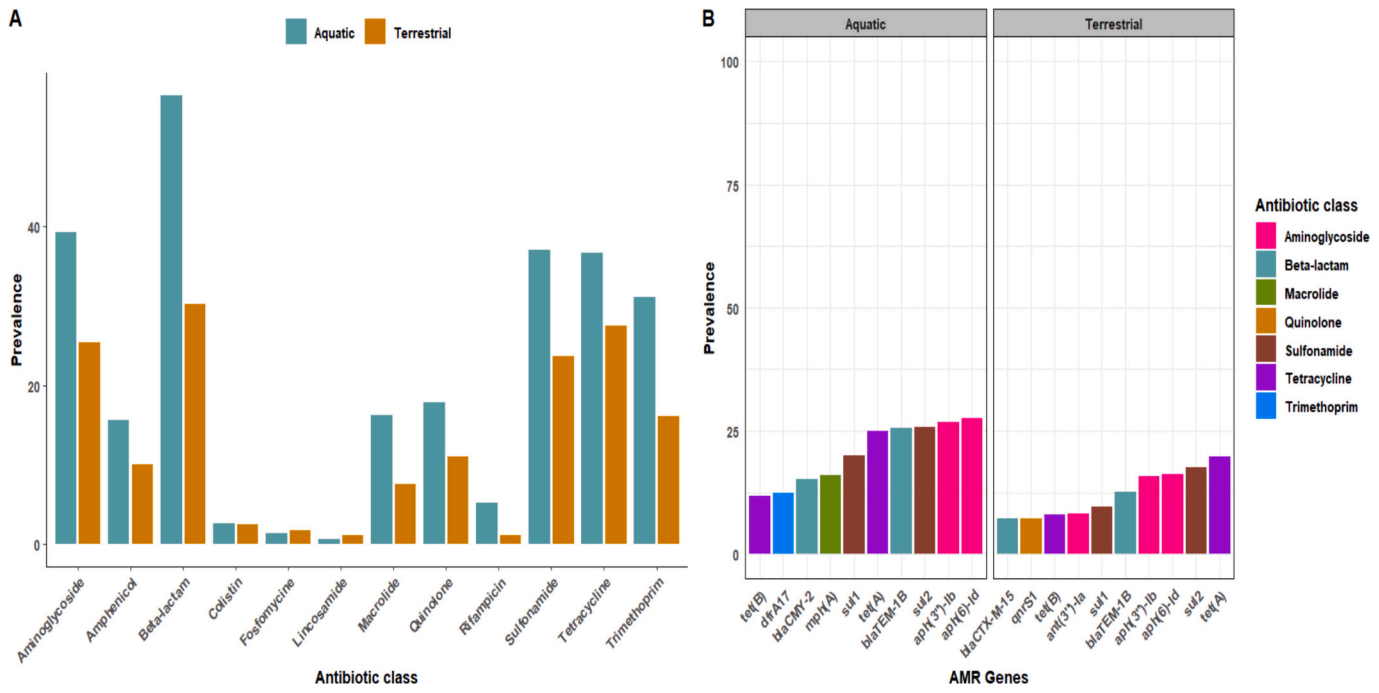


Fig. 4. Distribution of AMR per habitat. A: Barplot showing the prevalence of resistance to antibiotic class per habitat. B: Barplot showing the ARGs having the highest relative abundance per habitats colored by antibiotic class.

animals was for beta-lactams with a prevalence of 56.5 % in aquatic and 30.2 % in terrestrial animals (Fig. 4A).

The most abundant ARGs detected in aquatic animals were *aph(6)-Id* (27.5 %) and *aph(3″)-Ib* (26.7 %), while *tet(A)* (19.8 %) and *sul2* (17.6 %) were abundant in terrestrial animals. In addition, *aph(6)-Id* and *aph (3″)-Ib* showed similar within-habitat prevalence (Fig. 4B). Wild animals in aquatic environment harbored more ARGs resistant to the three clinically important antibiotics than their counterparts in the terrestrial ecosystems. In terrestrial wild animals, $bla_{\text{NDM-5}}$ was the only carbapenem-resistant gene identified, whereas approximately ten ARGs resistant to carbapenem were detected in aquatic animals, with $bla_{\text{IMP-4}}$ being the most abundant (4.9 %). The prevalence of colistin-resistant genes (*mcr-1* and *mcr-9*) was higher in aquatic animals. Furthermore, the most frequently detected ESBL-resistant gene in aquatic and terrestrial animals was $bla_{\text{CTX-M-15}}$, with a prevalence of 7.7 % and 7.1 %, respectively (Fig. S5).

4. Discussion

Theoretically, since wild animals are not treated with antimicrobial agents, they should not be directly implicated in the selection and development of AMR. However, it has been suggested that they can act as reservoirs, “mixing pots” and disseminators of AMR, resulting in important bioindicators of the impact of human activities on the environment (Allen et al., 2010; Bonnedahl and Järhult, 2014; Wu et al., 2018). Most studies on AMR are focused on humans and livestock. There are few studies on AMR in *E. coli* isolated from wild animals, but most of them focus on a few species and in restricted areas. The current study is the first study that includes worldwide *E. coli* genomic data from different wild animal species and shows the distribution of AMR across geographic regions, host categories and habitats of wild animals.

Overall, the genomes recovered from public databases showed a higher level of resistance to beta-lactams, aminoglycosides, tetracyclines, sulfonamides, trimethoprim and quinolones, antibiotics most commonly used in livestock and/or humans (Browne et al., 2021; Caneschi et al., 2023; Van Boeckel et al., 2017). Our finding shows a potential link between the occurrence of AMR in wild animals and the misuse or overuse of antimicrobials in humans and livestock. Even though the highest level of resistance was detected in beta-lactams, the most dominant ARG detected was tetracycline resistant gene *tet(A)*. This was due to the detection of a higher number of ARGs resistant to beta-lactam, as 58 (36.5 %) of the total 159 ARGs detected were beta-lactam-resistant genes (Table S2).

The present study revealed that the number of ARGs per genome varies across continents. The highest ARG per genome was detected in Asia, Africa and South and Central America, respectively, in decreasing order. However, this result should be interpreted with caution, given differences in health regulations and the use of antibiotics between countries in the same continent. This result was in line with the findings of Pursey et al. (2023), who reported high ARG carriage in most parts of Asia, Northern Africa, and South America using *E. coli* genomes from humans, domestic animals and wild animals. The high burden of AMR in these regions could be associated with poor hygiene and sanitation, limited access to quality healthcare, and the lack of regulations on the use of antibiotics in humans, animals and crops (Sulis et al., 2022). This coupled with poor waste management in some of these regions could be associated with the high prevalence of AMR in wild animals.

For instance, it is estimated that about 92 % of wastewater is discharged untreated in low-income countries, while only 30 % is released untreated in high-income countries (Khalid et al., 2018). Furthermore, the higher burden of AMR could be associated with the extensive livestock production system and the low biosecurity measures in those regions, which leads to the close interaction between livestock and wildlife (Jori et al., 2021).

According to Mulchandani et al. (2023) and Tiseo et al. (2020), antibiotic consumption in food-producing animals was and is expected to remain high in Asia, South America, Europe, North America, Africa and Oceania in decreasing order. The prevalence of ARGs in wild animals in the present study mirrors this finding, except for the fact that Africa and Oceania had lower antibiotic use but higher ARGs than Europe and North America. On the contrary, a higher rate of antibiotic use in humans was seen in North America and Europe, while the lowest rate was observed in sub-Saharan Africa and most parts of Asia (Browne et al., 2021). However, the antibiotic use data both in humans and

animals may not show the real situation as most countries, especially LMICs, do not report their data. For instance, the estimation of antibiotic consumption in food animals was based on reports from only 42 countries worldwide (Mulchandani et al., 2023).

Genomes from Asia harbored a remarkably higher *mcr-1* gene than those in other continents. This could be due to the high consumption of colistin in some Asian countries, especially China, the leading consumer and producer of colistin worldwide (Shen et al., 2016), until its ban in 2017. However, this variation might be also be attributed to sequencing bias, as many of the genomes may have been derived from studies specifically focusing on this gene. Furthermore, the higher detection rate of $bla_{\text{CTX-M-15}}$ in Africa is likely influenced by such targeted sequencing efforts. Given the limited accessibility and sequencing capacity in the region, most genomes from the region are more likely to be targeted.

In the present study, the distribution of ARGs also varies among different wild animal host groups. Wild birds had a higher ARGs per genome than wild mammals. This finding was consistent with previous phenotypic-based studies. Smith et al. (2014) found higher AMR bacteria in wild birds than in wild mammals. Genomes from migratory birds harbored a high number of $bla_{\text{CTX-M}}$ genes (ESBLs), mainly $bla_{\text{CTX-M-15}}$ genes. Although lower than in non-migratory birds, the higher ARGs in migratory birds could be associated with their migratory pattern, which magnifies the acquisition and dissemination of AMR over long distances (Bauer and Hoye, 2014; Stedt et al., 2014).

Non-migratory birds had a higher ARGs per genome and showed a relatively higher level of resistance for most classes of antibiotics except aminoglycoside, beta-lactam, sulfonamide and tetracycline. In addition, genomes from non-migratory birds had a higher level of resistance to critically important antibiotics including rifampicin and quinolone and harbored a higher prevalence of colistin (*mcr-1* and *mr-9*) and carbapenem (mainly $bla_{\text{IMP-4}}$) resistant genes. Non-migratory birds may not travel long distances but have the potential to spread AMR from hotspot areas to vulnerable populations (Arnold et al., 2016).

Wild carnivores had the highest ARGs per genome and a higher prevalence rate for all classes of antibiotics than any other wild mammals (ungulates, bats and other mammals). It has been reported that wild carnivores can acquire AMR bacteria in several ways; however, exposure to resistant isolates from anthropogenic sources and domestic animals is the main means of acquisition (Osińska et al., 2020; Sherley et al., 2000). Therefore, the result of the current study could be associated with the impact of anthropogenic activity in contaminating the environment, given carnivores' synanthropization behavior and large territorial coverage (Osińska et al., 2020).

Analysis of the ARGs distribution among habitats of wild animals showed that aquatic animals had a higher number of ARGs per isolate and a higher level of resistance to all classes of antibiotics, except fosfomycine and lincosamide, than those living in the terrestrial environment. Besides, the number of ARGs associated with resistant to carbapenem and ESBL genes were higher in aquatic animals. This finding supports the hypothesis that proximity to water bodies increases the likelihood of carrying AMR *E. coli* and is in line with the findings of other phenotype-based studies. For instance, it has been reported that wild small mammals living in a coastal area had a higher carriage rate and abundance of AMR *E. coli* than their inland counterparts (Furness et al., 2017). In addition, Jobbins and Alexander (2015) found that water-associated species (wild animals living or foraging in the aquatic environment) had a higher level of antibiotic resistance than non-water-associated species.

Most studies on the role of aquatic ecosystems in the selection of ARGs and AMR bacteria focus on aquaculture, as antibiotics are commonly used in this sector (Cabello et al., 2013). However, the present study showed that free-living wild animals living in the aquatic ecosystem could also play a role in the spread of AMR through water bodies. The higher burden of ARGs in aquatic wild animals could indicate the impact of human activities on the aquatic environment, as wastes from humans and livestock production can enter the waterways through runoff and sewage breaks. On the other hand, given that the contaminated water could be used for agricultural practices including crop production and aquaculture, it could be a further source of AMR for humans and domestic animals.

4.1. Limitations of the study

The present study comes with limitations. First, a substantial proportion of the genomes (over 60 %) originate from just two countries, the United States of America and Australia, while countries from Asia, Africa and South and Central America are represented by 8.2 % genomes. This uneven representation may introduce bias when interpreting the number of ARGs per isolate. Another limitation was the lack of standardized metadata, which led to the removal of those genomes from the analysis and could underestimate the burden of AMR in wild animals.

5. Conclusions

Overall, the burden of AMR in wildlife was higher in Asia, Africa and South and Central America. This finding highlights the impact of antibiotic misuse or overuse and other anthropogenic activities, like poor management of waste, on the occurrence of AMR in the environment. In addition, wild birds and carnivores harbored a higher number of ARGs per isolate and had the highest level of AMR to antibiotics widely used in humans and animals, as well as critically important or last-line antibiotics. In contrast, the lowest level of AMR, with barely 1 ARG per genome, was detected in bats and ungulates. Wild birds, mainly non-migratory birds, showed the highest prevalence of colistin-resistant genes. Furthermore, aquatic wild animals had a higher level of resistance to most classes of antibiotics and a higher number of ARGs per genome than terrestrial wild animals. Therefore, given the higher burden of AMR and the fact that they live in a diverse ecosystem, wild birds (migratory and non-migratory), carnivores, and all wild animals living in the aquatic environment could be good sentinels for the study of AMR pressure in wild animals and the environment.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2025.179742.

CRediT authorship contribution statement

Andnet Yirga Assefa: Writing – review & editing, Writing – original draft, Visualization, Software, Methodology, Investigation, Formal analysis, Data curation. **Biel Garcias:** Writing – review & editing, Supervision, Methodology, Formal analysis. **Evangelos Mourkas:** Writing – review & editing. **Rafael A. Molina-López:** Writing – review & editing. **Laila Darwich:** Writing – review & editing, Writing – original draft, Validation, Supervision, Methodology, Conceptualization.

Declaration of competing interest

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.

Acknowledgements

Andnet Yirga Assefa was supported by a scholarship from Erasmus Mundus Joint Master's Degree in Infectious Diseases and One Health Program (IDOH+). Biel Garcias was supported by the Departament de Recerca i Universitats de la Generalitat de Catalunya (FI-SDUR 2020-00376).

Data availability

Data will be made available on request.

References

Aerts, M., Battisti, A., Hendriksen, R., Kempf, I., Teale, C., Tenhagen, B.A., Veldman, K., Wasyl, D., Guerra, B., Liébana, E., Thomas-López, D., Belœil, P.A., 2019. Technical specifications on harmonised monitoring of antimicrobial resistance in zoonotic and indicator bacteria from food-producing animals and food. EFSA J. 17 (6). https://doi.org/10.2903/j.efsa.2019.5709.

Allen, H.K., Donato, J., Wang, H.H., Cloud-Hansen, K.A., Davies, J., Handelsman, J., 2010. Call of the wild: antibiotic resistance genes in natural environments. Nat. Rev. Microbiol. 8 (4). https://doi.org/10.1038/nrmicro2312.

Anjum, M.F., Schmitt, H., Börjesson, S., Berendonk, T.U., Donner, E., Stehling, E.G., Boerlin, P., Topp, E., Jardine, C., Li, X., Li, B., Dolejska, M., Madec, J.Y., Dagot, C., Guenther, S., Walsh, F., Villa, L., Veldman, K., Sunde, M., Pedersen, K., 2021. The potential of using *E. coli* as an indicator for the surveillance of antimicrobial resistance (AMR) in the environment. Curr. Opin. Microbiol. 64. https://doi.org/10.1016/j.mib.2021.09.011.

Arnold, K.E., Williams, N.J., Bennett, M., 2016. "Disperse abroad in the land": the role of wildlife in the dissemination of antimicrobial resistance. Biol. Lett. 12 (8). https://doi.org/10.1098/rsbl.2016.0137.

Atterby, C., Börjesson, S., Ny, S., Järhult, J.D., Byfors, S., Bonnedahl, J., 2017. ESBL-producing Escherichia coli in Swedish gulls—a case of environmental pollution from humans? PLoS One 12 (12). https://doi.org/10.1371/journal.pone.0190380.

Bauer, S., Hoye, B.J., 2014. Migratory animals couple biodiversity and ecosystem functioning worldwide. Science 344 (6179). https://doi.org/10.1126/science.1242552.

Blanco-Peña, K., Esperón, F., Torres-Mejía, A.M., De la Torre, A., de la Cruz, E., Jiménez-Soto, M., 2017. Antimicrobial resistance genes in pigeons from public parks in Costa Rica. Zoonoses Public Health 64 (7), e23–e30. https://doi.org/10.1111/zph.12340.

Bonnedahl, J., Järhult, J.D., 2014. Antibiotic resistance in wild birds. Ups. J. Med. Sci. 119 (2). https://doi.org/10.3109/03009734.2014.905663.

Bortolaia, V., Kaas, R.S., Ruppe, E., Roberts, M.C., Schwarz, S., Cattoir, V., Philippon, A., Allesoe, R.L., Rebelo, A.R., Florensa, A.F., Fagelhauer, L., Chakraborty, T., Neumann, B., Werner, G., Bender, J.K., Stingl, K., Nguyen, M., Coppens, J., Xavier, B. B., Aarestrup, F.M., 2020. Resfinder 4.0 for predictions of phenotypes from genotypes. J. Antimicrob. Chemother. 75 (12). https://doi.org/10.1093/jac/dkaa345.

Browne, A.J., Chipeta, M.G., Haines-Woodhouse, G., Kumaran, E.P.A., Hamadani, B.H. K., Zaraa, S., Henry, N.J., Deshpande, A., Reiner, R.C., Day, N.P.J., Lopez, A.D., Dunachie, S., Moore, C.E., Stergachis, A., Hay, S.I., Dolecek, C., 2021. Global antibiotic consumption and usage in humans, 2000–18: a spatial modelling study. Lancet Planet. Health 5 (12). https://doi.org/10.1016/S2542-5196(21)00280-1.

Cabello, F.C., Godfrey, H.P., Tomova, A., Ivanova, L., Dölz, H., Millanao, A., Buschmann, A.H., 2013. Antimicrobial use in aquaculture re-examined: its relevance to antimicrobial resistance and to animal and human health. Environ. Microbiol. 15 (7). https://doi.org/10.1111/1462-2920.12134.

Caneschi, A., Bardhi, A., Barbarossa, A., Zaghini, A., 2023. The use of antibiotics and antimicrobial resistance in veterinary medicine, a complex phenomenon: a narrative review. Antibiotics 12 (3). https://doi.org/10.3390/antibiotics12030487.

Carroll, D., Wang, J., Fanning, S., McMahon, B.J., 2015. Antimicrobial resistance in wildlife: implications for public health. Zoonoses Public Health 62 (7), 534–542. https://doi.org/10.1111/zph.12182.

Darwich, L., Vidal, A., Seminati, C., Albamonte, A., Casado, A., López, F., Molina-López, R.A., Migura-Garcia, L., 2019. High prevalence and diversity of extended-spectrum β-lactamase and emergence of OXA-48 producing Enterobacterales in wildlife in Catalonia. PLoS One 14 (8). https://doi.org/10.1371/journal.pone.0210686.

Del Hoyo, J., 2020. All the Birds of the World. Lynx Edicions.

Delgado-Blas, J.F., Ovejero, C.M., David, S., Montero, N., Calero-Caceres, W., Garcillan-Barcia, M.P., de la Cruz, F., Muniesa, M., Aanensen, D.M., Gonzalez-Zorn, B., 2021. Population genomics and antimicrobial resistance dynamics of Escherichia coli in wastewater and river environments. Communications Biology 4 (1). https://doi.org/10.1038/s42003-021-01949-x.

Dias, D., Torres, R.T., Kronvall, G., Fonseca, C., Mendo, S., Caetano, T., 2015. Assessment of antibiotic resistance of Escherichia coli isolates and screening of Salmonella spp. in wild ungulates from Portugal. Res. Microbiol. 166 (7). https://doi.org/10.1016/j.resmic.2015.03.006.

Furness, L.E., Campbell, A., Zhang, L., Gaze, W.H., McDonald, R.A., 2017. Wild small mammals as sentinels for the environmental transmission of antimicrobial resistance. Environ. Res. 154. https://doi.org/10.1016/j.envres.2016.12.014.

Guenther, S., Ewers, C., Wieler, L.H., 2011. Extended-spectrum beta-lactamases producing E. coli in wildlife, yet another form of environmental pollution? Front. Microbiol. 2, 246.

Huijbers, P.M.C., Larsson, D.G.J., Flach, C.F., 2020. Surveillance of antibiotic resistant Escherichia coli in human populations through urban wastewater in ten European countries. Environ. Pollut. 261. https://doi.org/10.1016/j.envpol.2020.114200.

Hutinel, M., Huijbers, P.M.C., Fick, J., Åhrén, C., Larsson, D.G.J., Flach, C.F., 2019. Population-level surveillance of antibiotic resistance in Escherichia coli through sewage analysis. Eurosurveillance 24 (37). https://doi.org/10.2807/1560-7917.ES.2019.24.37.1800497.

Jiang, L., Hu, X., Xu, T., Zhang, H., Sheng, D., Yin, D., 2013. Prevalence of antibiotic resistance genes and their relationship with antibiotics in the Huangpu River and the drinking water sources, Shanghai, China. Sci. Total Environ. 458, 267–272. https://doi.org/10.1016/j.scitotenv.2013.04.038.

Jobbins, S.E., Alexander, K.A., 2015. From whence they came—antibiotic-resistant *Escherichia coli* in African wildlife. J. Wildl. Dis. 51 (4). https://doi.org/10.7589/2014-11-257.

Jones, K.E., Patel, N.G., Levy, M.A., Storeygard, A., Balk, D., Gittleman, J.L., Daszak, P., 2008. Global trends in emerging infectious diseases. Nature 451 (7181), 990–993. https://doi.org/10.1038/nature06536.

Jori, F., Hernandez-Jover, M., Magouras, I., Dürr, S., Brookes, V.J., 2021. Wildlife-livestock interactions in animal production systems: what are the biosecurity and health implications? Anim. Front. 11 (5). https://doi.org/10.1093/af/vfab045.

Khalid, S., Shahid, M., Natasha, Bibi, I., Sarwar, T., Shah, A.H., Niazi, N.K., 2018. A review of environmental contamination and health risk assessment of wastewater use for crop irrigation with a focus on low and high-income countries. Int. J. Environ. Res. Public Health 15 (5). https://doi.org/10.3390/ijerph15050895.

Lincoln, F.C., Peterson, S.R., Zimmerman, J.L., 1998. Migration of Birds Circular 16.

Lipworth, S., Crook, D., Walker, A.S., Peto, T., Stoesser, N., 2024. Exploring uncatalogued genetic variation in antimicrobial resistance gene families in Escherichia coli: an observational analysis. The Lancet Microbe 5 (11). https://doi.org/10.1016/S2666-5247(24)00152-6.

Łopucki, R., Stępień-Pyśniak, D., Christensen, H., Kubiński, K., Lenarczyk, E., Martinez-de-Tejada, G., Masłyk, M., 2024. Interspecies transmission of antimicrobial-resistant bacteria between wild birds and mammals in urban environment. Vet. Microbiol., 110130 https://doi.org/10.1016/j.vetmic.2024.110130.

Miller, S.A., Ferreira, J.P., LeJeune, J.T., 2022. Antimicrobial use and resistance in plant agriculture: a one health perspective. Agriculture 12 (2), 289. https://doi.org/10.3390/agriculture12020289.

Mulchandani, R., Wang, Y., Gilbert, M., Van Boeckel, T.P., 2023. Global trends in antimicrobial use in food-producing animals: 2020 to 2030. PLoS Glob. Public Health 3 (2). https://doi.org/10.1371/journal.pgph.0001305.

Murray, C.G., Hamilton, A.J., 2010. Perspectives on wastewater treatment wetlands and waterbird conservation. J. Appl. Ecol. 47 (5), 976–985. https://doi.org/10.1111/j.1365-2664.2010.01853.x.

Murray, C.J., Ikuta, K.S., Sharara, F., Swetschinski, L., Robles Aguilar, G., Gray, A., Han, C., Bisignano, C., Rao, P., Wool, E., Johnson, S.C., Browne, A.J., Chipeta, M.G., Fell, F., Hackett, S., Haines-Woodhouse, G., Kashef Hamadani, B.H., Kumaran, E.A. P., McManigal, B., Naghavi, M., 2022. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. Lancet 399 (10325), 629–655. https://doi.org/10.1016/S0140-6736(21)02724-0.

Olaru, I.D., Walther, B., Schaumburg, F., 2023. Zoonotic sources and the spread of antimicrobial resistance from the perspective of low and middle-income countries. Infect. Dis. Poverty 12 (03), 73–87. https://doi.org/10.1186/s40249-023-01113-z.

O'Neil, J., 2014. Review on antibiotic resisitance. In: Antimicrobial Resistance: Tackling a Crisis for the Health and Wealth of Nations. Health and Wealth Nations (December).

Oravcova, V., Ghosh, A., Zurek, L., Bardon, J., Guenther, S., Cizek, A., Literak, I., 2013. Vancomycin-resistant enterococci in rooks (Corvus frugilegus) wintering throughout Europe. Environ. Microbiol. 15 (2), 548–556. https://doi.org/10.1111/1462-2920.12002.

Osińska, M., Nowakiewicz, A., Zięba, P., Gnat, S., Łagowski, D., Trościańczyk, A., 2020. Wildlife carnivorous mammals as a specific mirror of environmental contamination with multidrug-resistant Escherichia coli strains in Poland. Microb. Drug Resist. 26 (9). https://doi.org/10.1089/mdr.2019.0480.

Pires, J., Huisman, J.S., Bonhoeffer, S., Van Boeckel, T.P., 2021. Multidrug resistance dynamics in *Salmonella* in food animals in the United States: an analysis of genomes from public databases. Microbiol. Spectr. 9 (2). https://doi.org/10.1128/spectrum.00495-21.

Pires, J., Huisman, J.S., Bonhoeffer, S., Van Boeckel, T.P., 2022. Increase in antimicrobial resistance in Escherichia coli in food animals between 1980 and 2018 assessed using genomes from public databases. J. Antimicrob. Chemother. 77 (3). https://doi.org/10.1093/jac/dkab451.

Pursey, E., Dimitriu, T., Gaze, W.H., Westra, E.R., Van Houte, S., 2023. The distribution of antimicrobial resistance genes across phylogroup, host species and geography in 16,000 publicly-available E. coli genomes. MedRxiv. https://doi.org/10.1101/2022.08.05.22278465.

R Core Team, 2024. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Schaefer, A.M., Goldstein, J.D., Reif, J.S., Fair, P.A., Bossart, G.D., 2009. Antibiotic-resistant organisms cultured from Atlantic bottlenose dolphins (Tursiops truncatus) inhabiting estuarine waters of Charleston, SC and Indian River Lagoon, FL. Ecohealth 6, 33–41. https://doi.org/10.1007/s10393-009-0221-5.

Schar, D., Klein, E.Y., Laxminarayan, R., Gilbert, M., Van Boeckel, T.P., 2020. Global trends in antimicrobial use in aquaculture. Sci. Rep. 10 (1), 21878. https://doi.org/10.1038/s41598-020-78849-3.

Shen, Z., Wang, Y., Shen, Y., Shen, J., Wu, C., 2016. Early emergence of *mcr-1* in *Escherichia coli* from food-producing animals. Lancet Infect. Dis. 16 (3). https://doi.org/10.1016/S1473-3099(16)00061-X.

Sherley, M., Gordon, D.M., Collignon, P.J., 2000. Variations in antibiotic resistance profile in Enterobacteriaceae isolated from wild Australian mammals. Environ. Microbiol. 2 (6). https://doi.org/10.1046/j.1462-2920.2000.00145.x.

Smith, S., Wang, J., Fanning, S., McMahon, B.J., 2014. Antimicrobial resistant bacteria in wild mammals and birds: a coincidence or cause for concern? Ir. Vet. J. 67 (1). https://doi.org/10.1186/2046-0481-67-8.

Spina, F., Baillie, S.R., Bairlein, F., Ambrosini, R., Fattorini, N., Fiedler, W., Franks, S., Funghi, C., Thorup, K., 2023. Eurasian-African bird migration atlas executive summary. https://migrationatlas.org.

Stedt, J., Bonnedahl, J., Hernandez, J., McMahon, B.J., Hasan, B., Olsen, B., Drobni, M., Waldenström, J., 2014. Antibiotic resistance patterns in Escherichia coli from gulls in nine European countries. Infection Ecology & Epidemiology 4 (1). https://doi.org/10.3402/iee.v4.21565.

Sulis, G., Sayood, S., Gandra, S., 2022. Antimicrobial resistance in low- and middle-income countries: current status and future directions. Expert Rev. Anti-Infect. Ther. 20 (2). https://doi.org/10.1080/14787210.2021.1951705.

Swift, B.M., Bennett, M., Waller, K., Dodd, C., Murray, A., Gomes, R.L., Arnold, K.E., 2019. Anthropogenic environmental drivers of antimicrobial resistance in wildlife. Sci. Total Environ. 649, 12–20. https://doi.org/10.1016/j.scitotenv.2018.08.180.

Tiseo, K., Huber, L., Gilbert, M., Robinson, T.P., Van Boeckel, T.P., 2020. Global trends in antimicrobial use in food animals from 2017 to 2030. Antibiotics 9 (12). https://doi.org/10.3390/antibiotics9120918.

UNEP-WCMC, 2024. State of the world's migratory species. www.unep-wcmc.org.

Van Boeckel, T.P., Brower, C., Gilbert, M., Grenfell, B.T., Levin, S.A., Robinson, T.P., Laxminarayan, R., 2015. Global trends in antimicrobial use in food animals. Proc. Natl. Acad. Sci. 112 (18), 5649–5654. https://doi.org/10.1073/pnas.1503141112.

Van Boeckel, T.P., Glennon, E.E., Chen, D., Gilbert, M., Robinson, T.P., Grenfell, B.T., Levin, S.A., Bonhoeffer, S., Laxminarayan, R., 2017. Reducing antimicrobial use in food animals. Science 357 (6358). https://doi.org/10.1126/science.aao1495.

Van Boeckel, T.P., Pires, J., Silvester, R., Zhao, C., Song, J., Criscuolo, N.G., Gilbert, M., Bonhoeffer, S., Laxminarayan, R., 2019. Global trends in antimicrobial resistance in animals in low- and middle-income countries. Science 365 (6459). https://doi.org/10.1126/science.aaw1944.

Vieira, A.D.A., Piccoli, B.C., y Castro, T.R., Casarin, B.C., Tessele, L.F., Martins, R.C.R., Trindade, P.D.A., 2023. Pipeline validation for the identification of antimicrobial-resistant genes in carbapenem-resistant Klebsiella pneumoniae. Sci. Rep. 13 (1). https://doi.org/10.1038/s41598-023-42154-6.

Vittecoq, M., Godreuil, S., Prugnolle, F., Durand, P., Brazier, L., Renaud, N., Arnal, A., Aberkane, S., Jean-Pierre, H., Gauthier-Clerc, M., Thomas, F., Renaud, F., 2016. Review: antimicrobial resistance in wildlife. J. Appl. Ecol. 53 (2), 519–529. https://doi.org/10.1111/1365-2664.12596 (Blackwell Publishing Ltd.).

Wallace, C.C., Yund, P.O., Ford, T.E., Matassa, K.A., Bass, A.L., 2013. Increase in antimicrobial resistance in bacteria isolated from stranded marine mammals of the Northwest Atlantic. EcoHealth 10, 201–210. https://doi.org/10.1007/s10393-013-0842-6.

World Bank, 2017. Drug-resistant Infections: A Threat to Our Economic Future. World Bank, Washington, DC. License: Creative Commons Attribution CC BY 3.0 IGO. www.worldbank.org.

Wu, J., Huang, Y., Rao, D., Zhang, Y., Yang, K., 2018. Evidence for environmental dissemination of antibiotic resistance mediated by wild birds. Front. Microbiol. 9 (APR). https://doi.org/10.3389/fmicb.2018.00745.