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Antimicrobial Resistance Profiles and Metallo-*β*-Lactamase Genes (*bla*_NDM, *bla*_IMP) in Enterobacteriaceae Isolated from Wild Bats in Burkina Faso

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Abstract

The global resurgence of infectious diseases has renewed attention on bats, recognized reservoirs of diverse pathogens, including antibiotic-resistant bacteria that pose significant public health threats. This study provides the first comprehensive assessment in Burkina Faso of the antibiotic resistance profiles of Enterobacteriaceae isolated from bats, focusing on three key antibiotic families: *β*-lactams, third-generation cephalosporins, and carbapenems. From December 2020 to September 2021, bats were captured using mist nets across six locations in Burkina Faso. Oropharyngeal and rectal swabs were collected and processed for bacterial isolation, identification, and antimicrobial susceptibility testing, followed by molecular screening for *bla*_IMP and *bla*_NDM metallo-*β*-lactamase genes. Among 204 bats sampled, 84 tested positive for at least one enterobacterial species, corresponding to a prevalence of 41.17%. The infected individuals belonged to six genera and nine species, comprising one frugivorous species (*Epomophorus gambianus*) and eight insectivorous species (*Hypsideros caffer*, *Hypsideros jonesi*, *Mops condylurus*, *Mops midas*, *Mops pumilus*, *Pipistrellus nanulus*, *Rhinolophus alcyone*, and *Scotophilus leuco-*

gaster). A total of 29 Enterobacteriaceae species were isolated and tested. Resistance to third-generation cephalosporins was observed in 35% of isolates, to *β*-lactams (amoxicillin-clavulanic acid) in 46%, and to carbapenems in 2.4%. Ceftriaxone (CRO) resistance was detected in 11 bacterial species and reached 100% in Salmonella Typhi and Salmonella Paratyphi A. While resistance profiles to ceftriaxone varied across bat species, no significant differences were detected for amoxicillin-clavulanic acid. Bacterial resistance patterns were not associated with bat sex. Molecular analyses revealed a high prevalence of the *bla*_NDM gene (96.7%) compared to *bla*_IMP (3.3%) among resistant isolates. These findings highlight bats as potential reservoirs of multidrug-resistant enterobacteria in West Africa, emphasizing the need for integrated One Health surveillance to better understand wildlife-human transmission risks.

Keywords

Bats, Chiroptera, Enterobacteriaceae, Wildlife, Antimicrobial Resistance, *bla*_NDM, *bla*_IMP, Burkina Faso, West Africa

1. Introduction

Antibiotics resistance is one of today's major public health problems responsible for thousands of human fatalities annually [1]-[3].

However, the contribution of wildlife to the spread of antibiotics resistance among different hosts and ecosystems is significant [4] [5]. This wildlife is generally not exposed to clinically used antibiotics [6]. However, wild animals can become infected with antibiotic-resistant bacteria through contact with humans or the environment [7]. Thus, once these animals are contaminated, they can in turn become reservoirs and vectors for the transmission of highly pathogenic bacteria to humans [1] [8] [9]. Indeed, Enterobacteria seem to be more specifically adapted to humans or animals; some are responsible for human infections and sometimes very severe (typhoid fever, bacillary dysentery) in view of their continued resistance and virulence. One of the most striking developments in human and animal antibiotic resistance in recent decades has been the resistance of Enterobacteriaceae to the latest generation of beta-lactams and cephalosporins [10]. This resistance is mainly ensured by the production of extended-spectrum beta-lactamases (ESBL) and to a lesser extent plasmid cephalosporinases (AmpC). These enzymes confer high resistance to most therapeutic beta-lactams (with the notable exception of carbapenems). Their genes, mainly located on plasmids, spread easily between bacteria, thus increasing their resistance to antibiotics [11]-[13]. IMP, which stands for "active against imipenem" or "imipenemase," is a class B bacterial ambler metabolo-bêta-lactamases that are often found in Pseudomonas aeruginosa but may also be found in enterobacteriaceae (also known as bacteria). There are at least 52 distinct variants in this diverse category of metallo-beta-lactamases (Logan and Wein-

stein, 2017). The New Delhi Metallo-β-lactamase-1 (NDM-1) presents a challenge in the treatment of bacterial infections by providing resistance to carbapenem antibiotics [14].

Indeed, antibiotic-resistant strains of Enterobacteriaceae have already been isolated from wildlife and, most notably, from bats [15]-[18]. Specimens of *Escherichia coli* bacteria isolated from insectivorous bats showed 15% resistance to MCA (amoxicillin + clavulanic acid) in surveys [19]. Since we share a common environment between animals and humans, bats usually roost near human dwellings; they are carriers of many microorganisms that are pathogenic to humans and animals [7]. Bat bacterium knowledge is still very scarce, but a few studies have indicated that bats are hosts of antimicrobial resistance and play an important role in the dispersion of resistance in the environment [15]. Worldwide, 32 studies on bacterial resistance in bats have been conducted between 1985 and 2021, including nine in Africa [16]. Among those studies that have already addressed the resistance of bacteria isolated from bats in Africa are: Antimicrobial resistance in *Escherichia coli* isolates from frugivorous (*Eidolon helvum*) and insectivorous (*Nycteris hispida*) bats in Nigeria [19]. However, in Burkina Faso, studies on bat and their pathogens are very fragmentary [20] [21] and no research has yet been conducted on the antibiotic resistance of bacteria in bats in Burkina Faso.

The objective of this survey is to evaluate the resistance of enterobacteria isolated from bats to three families of antibiotics and characterize *blaIMP* and *blaNDM* resistance genes in Enterobacteriaceae isolates from bats.

2. Materials and Methods

2.1. Ethics Statement

The code of ethics was obtained from the Ethics Committee on Animal Experimentation of the Joseph KI-ZERBO University (permit no. CODE AGREEMENT: CEEA-UJKZ/2021-04).

The authorization to collect bats was obtained through the implementation of the framework cooperation agreement between Joseph Ki-ZERBO University and the Ministry of the Environment, Green Economy and Climate Change (MEECVCC) (permit no. 21-091 MEECVCC/SG/DGEF/DFRC).

2.2. Study Area

This survey was conducted in six (06) cities in Burkina Faso and each city had several sampling points. The central region had seven sampling points as shown in **Figure 1**. These sites were chosen based on the presence of waterways, forest galleries, caves and abandoned houses, which are the bats' preferred locations.

2.3. Sample Collection

Bats were captured using mist nets from December 2020 to September 2021. Mist nets were placed in several places (e.g., dwellings, vegetation galleries, along water points). Care was taken to free the bats, and particular attention was paid to wing

clearances because bats enter the nets with their wings spread and then fold them back up. The nets were opened at nightfall from 6 pm to 5 am depending on the activity of the bats.

Each individual was placed in a porous cotton bag and kept until the time of sampling. The risks of contamination were minimized as much as possible by using protective suits during the capture and handling of the bats. In order to identify bats, we determined weight, sex, and forearm length. Weight was measured with the pressure scale. Observation of the presence of male or female genitalia was used to determine the sex of the animal [22]. Bats were identified using the guide and identification key [23]-[25].

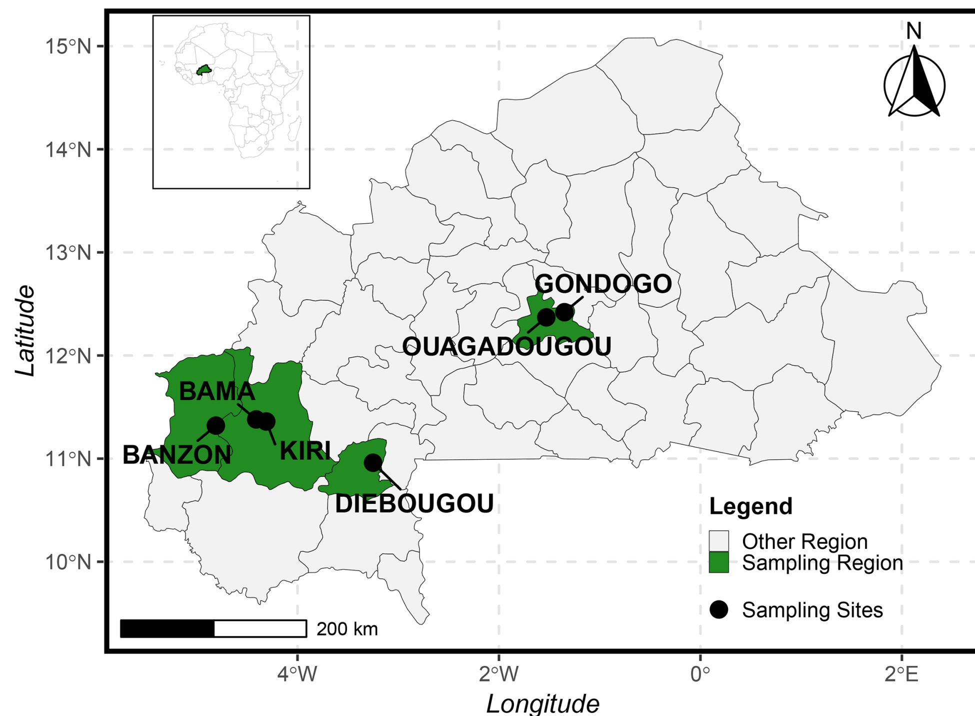

Figure 1. Site of capture of bats with the abundance of bats captured on each site.

2.4. Microbiology Analyses

Sterile swabs were used to collect oral and rectal samples from bats under aseptic conditions. Each swab was removed from its packaging without allowing the tip to come into contact with any surface, including gloves. Oral samples were collected by gently rotating the swab inside the oral cavity and behind the tongue. Rectal samples were obtained by carefully inserting the swab tip into the anus and gently rotating it against the mucosal surface for two to four circular motions. Excess fecal material was removed by gentle shaking before placing the swab into a cryovial containing transport medium.

Bacterial isolation was performed using selective and differential media. Swabs were inoculated onto URIselect, Hektoen, and Salmonella-Shigella (SS) agar plates and incubated at 37˚C for 24 h. Suspected colonies were subcultured for purifica-

tion and identified using the API 20E system (bioMérieux, France).

Antimicrobial susceptibility testing was conducted using the disk diffusion method on Mueller-Hinton agar, in accordance with the guidelines of the Antibiogram Committee of the French Society of Microbiology (CA-SFM). The antibiotics tested included ceftriaxone (CRO; third-generation cephalosporin), amoxicillin-clavulanic acid (AMC; β-lactam), and imipenem (IMI; carbapenem).

A bacterial suspension was prepared from a pure colony in 5 mL of sterile physiological saline and adjusted to 0.5 McFarland turbidity. The suspension was evenly spread onto Mueller-Hinton agar plates (4 mm thickness). Antibiotic discs were aseptically placed on the agar surface at a minimum distance of 20 mm from each other. Plates were incubated at 37˚C for 18 - 24 h.

After incubation, inhibition zone diameters were measured using a calibrated ruler and interpreted according to CA-SFM breakpoints. Isolates were classified as susceptible (S), intermediate (I), or resistant (R). Detection of extended-spectrum β-lactamase (ESBL) production was performed using the double-disk synergy test, based on the interaction between clavulanic acid and third-generation cephalosporins [26].

Quality control for antimicrobial susceptibility testing was performed using reference strains, including *Escherichia coli* ATCC 25922 (susceptible control) and *Klebsiella pneumoniae* ATCC 700603 (ESBL-positive control), to validate test performance [13].

2.5. Molecular Analysis

For bacterial nucleic acid extraction, genomic DNA was extracted using the Sacace™ DNA-Sorb-B nucleic acid extraction kit (Version 2020, K-1-1/B/100) according to the manufacturer's instructions. Real-time polymerase chain reaction (qPCR) was performed to detect two resistance-associated genes, bla_NDM and bla_IMP, which encode New Delhi metallo-β-lactamase and IMP-type metallo-β-lactamase, respectively.

Each PCR run included appropriate quality controls. DNA from known positive strains was used as a positive control (*Escherichia coli* ATCC 25922), while sterile distilled water was used as a negative control in place of template DNA to detect potential contamination and ensure the reliability of the amplification results (**Table 1**).

Table 1. Representation of the different primers for real-time PCR.

Genes	Primers	Sequences	Size (bp)	References
*bla*IMP	For	5' CATGGTTTGGTGGTTCTTGT 3'	488	[27]
	Rev	5' ATAATTTGGCGGACTTTGGC 3'		
*bla*NDM	For	5' CAGCACACT TCCTATCTC 3'	292	[27]
	Rev	5' CCGCAACCATCCCCTCTT 3'		

2.6. Statistical Analyses

Statistical analyses were performed using R software (version 4.1.0). Data normality was assessed using the Shapiro-Wilk test and indicated that the variables were not normally distributed. Consequently, non-parametric statistical methods were applied.

Comparisons among groups were conducted using the Kruskal-Wallis rank-sum test for continuous variables. Associations between categorical variables were assessed using Pearson's Chi-squared test when expected cell frequencies were sufficient. For contingency tables with small sample sizes or expected counts below five, Fisher's Exact Test was applied to ensure statistical validity.

Proportions and statistical comparisons were calculated using the *tbl_summary* and *add_p* functions of the *gtsummary* package. Standardized Pearson residuals derived from Chi-squared tests were computed to identify cells contributing most strongly to significant associations and were visualized using the *assocplot* function from the *reprex* package.

All statistical tests were two-tailed, and a p-value < 0.05 was considered statistically significant.

3. Results

3.1. Bat Diversity and Bacterial Resistance Patterns to three Families of Antibiotics

Of a total of 204 bats captured, 84 bats were positive for the presence of at least one enterobacterium with a prevalence of 41.17%. The infected bats were composed of six genera and nine species, including one frugivore (*Epomophorus gambianus* (Ogilby, 1835)) and eight insectivores (*Hyposideros caffer* (Sundevall, 1846)), *Hyposideros jonesi* Hayman, 1947, *Mops condylurus* (A. Smith, 1833), *Mops midas* (Sundevall, 1843), *Mops pumilus* (Cretzschmar, 1830), *Pipistrellus nanulus* (Peters, 1868), *Rhinolophus alcyone* Temminck, 1853, *Scotophilus leucogaster* (Cretzschmar, 1826). Of the nine bat species, 29 species of enterobacteria were isolated and tested with three families of antibiotics (Table 2).

Table 2. The different bacterial species with their resistance to CRO, AMC, IMI antibiotics isolated from bat species

Bats	Bacteria species	ANTIBIOTIC RESISTANCE		
		CRO	AMC	IMI
Epomophorus gambianus	*Escherichia coli*	R	R	S
	Klebsiella oxytoca	R	R	S
	Citrobacter spp.	S	S	S
	Proteus spp.	S	S	S
	Escherichia coli	R	R	S
	Proteus mirabilis	S	R	S
	Salmonella paratyphi	R	R	S
	Salmonella Typhi	R	R	S

Continued

	Escherichia coli	S	S	S
	Enterobacter cloacea	S	S	S
	Citrobacter spp.	S	S	S
	Shigella spp.	R	R	S
	Salmonella spp.	R	R	S
	Klebsiella spp.	R	S	S
	Escherichia coli	R	S	S
	Proteus spp.	R	R	S
	Morganella morganii	S	S	S
	Shigella spp.	S	R	S
	Salmonella spp.	R	R	S
	Escherichia coli	S	R	S
	Klebsiella oxytoca	S	S	S
Mops midas	*Serratia* spp.	S	S	S
	Citrobacter spp.	S	S	S
	Citrobacter spp.	S	S	S
	Klebsiella spp.	S	S	S
	Citrobacter braakii	S	S	S
	Serratia liquefaciens	S	S	S
	Salmonella paratyphi	R	R	S
	Citrobacter spp.	S	R	S
	Citrobacter freundii	S	S	S
	Escherichia coli	S	R	S
	Citrobacter braakii	S	S	S
	Proteus vulgaris	S	S	S
	Escherichia coli	S	S	S
	Enterobacter sakazakii	S	S	S
	Enterobacter aerogenes	S	S	S
Mops condylurus	*Yersinia enterocolitica*	S	S	S
	Salmonella Typhi	R	R	S
	Proteus mirabilis	S	R	S
	Salmonella spp.	S	R	S
	Klebsiella spp.	R	R	S
	Yersinia spp.	S	S	S
	Hafnia alvei	S	S	S
	Escherichia coli	S	R	S
	Escherichia coli	S	S	S
	Klebsiella planticola	S	S	S
	Citrobacter spp.	S	S	S
	Enterobacter spp.	S	R	S

Continued

	Salmonella spp.	R	R	R
	Salmonella paratyphi A	R	R	R
	Proteus vulgaris	S	S	S
	Escherichia coli	R	S	S
	Enterobacter cloacea	S	S	S
	Escherichia coli	S	S	S
	Escherichia coli	S	S	S
	Salmonella spp.	S	R	S
	Serratia spp.	S	S	S
Mops pumilus	*Escherichia coli*	R	R	S
Scotophilus leucogaster	*Klebsiella* spp.	S	S	S
	Citrobacter spp.	S	S	S
	Escherichia coli	S	S	S
	Escherichia coli	S	S	S
	Klebsiella spp.	S	S	S
	Salmonella spp.	R	R	S
	Escherichia coli	S	R	S
	Escherichia coli	S	S	S
	Citrobacter spp.	S	S	S
	Klebsiella oxytoca	R	R	S
	Salmonella paratyphi	S	R	S
	Enterobacter spp.	S	S	S
	Escherichia coli	S	S	S
	Escherichia coli	R	R	S
	Escherichia coli	S	S	S
Hyposideros caffer	*Salmonella* spp.	S	R	S
	Escherichia coli	R	R	S
	Escherichia coli	R	R	S
Hyposideros jonesi	*Klebsiella* spp.	R	R	S
	Salmonella spp.	R	R	S
Pipistrellus nanulus	*Salmonella paratyphi A*	R	R	S
	Providencia rettgeri	S	S	S
	Enterobacter cloacea	S	S	S
Rhinolophus alcyone	*Escherichia coli*	R	R	S
	Escherichia coli	R	R	S
	Citrobacter spp.	R	R	S

Of all the bacteria isolated from bats in Burkina Faso, the majority are sensitive to third generation cephalosporins (CRO), betalactamines (AMC) and almost totally to carbapenems (IMI). There is no difference in resistance and susceptibility of the bacteria between AMC and CRO. The table below shows that 35% of the bac-

terial species are resistant to 3rd generation cephalosporins, 46% are resistant to betalactam (AMC) and 2.4% are resistant to carbapenems (**Figure 2**).

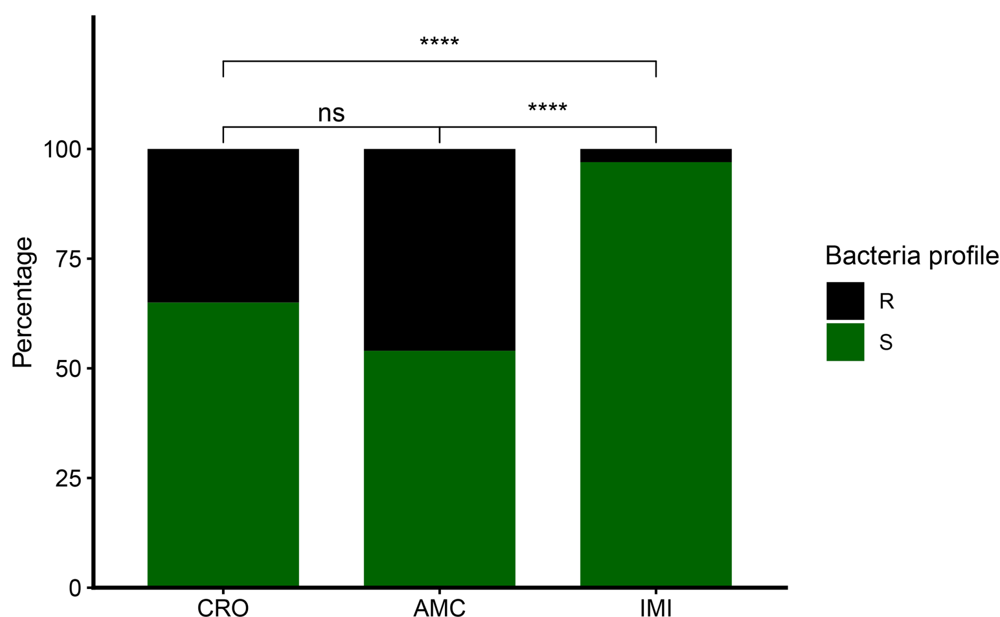

Figure 2. Percentage of bacterial profiles according to the three families of antibiotics.

Resistance to CRO was observed in 11 species of bacteria and was 100% in *Salmonella Typhi* and *Salmonella paratyphi* A. 13 species showed resistance with AMC and this resistance was 100% in six species, namely *Proteus mirabilis*, *Salmonella Typhi*, *Salmonella paratyphi*, *Salmonella paratyphi* A, *Salmonella* spp., and *Shigella* spp. With IMI, resistance was only observed in two species, *Salmonella* spp. and *Salmonella paratyphi* A and it was 100%. (**Table 3**). Thus, it is clear from **Figure 3** that two species of bacteria are resistant to all three families of antibiotics and these are *Salmonella* spp. and *Salmonella paratyphi* A. Two species are resistant only to CRO and 13 species only to AMC (**Table 3**).

Table 3. Abundance and percentage of resistance of bacterial species to the three antibiotic families.

Bacteria	CRO		AMC		IMI	
	R	S	R	S	R	S
Klebsiella oxytoca	2 (66, 66)	1 (33, 33)	2 (66, 66)	1 (33, 33)	0 (0)	3 (100)
Morganella morganii	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Acinobacter baumaani	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Acinobacter calcoaceticus,	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Acinotobacter spp.	1 (33, 33)	2 (66, 66)	1 (33, 33)	2 (66, 66)	0 (0)	3 (100)
Citrobacter braakii	0 (0)	2 (100)	0 (0)	2 (100)	0 (0)	2 (100)
Citrobacter freundii	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Citrobacter spp.	1 (11, 11)	8 (88, 88)	2 (22, 22)	7 (77, 77)	0 (0)	9 (100)
Enterobacter aerogenes	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)

Continued

Enterobacter cloacea	0 (0)	3 (100)	0 (0)	3 (100)	0 (0)	3 (100)
Enterobacter sakazakii	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Enterobacter spp.	0 (0)	2 (100)	1 (50)	1 (50)	0 (0)	2 (100)
Escherichia coli	10 (41, 66)	14 (58, 33)	12 (50)	12 (50)	0 (0)	24 (100)
Hafnia alvei	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Klebsiella planticola	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Klebsiella spp.	3 (50)	3 (50)	2 (33, 33)	4 (66, 66)	0 (0)	6 (100)
Proteus mirabilis	0 (0)	2 (100)	2 (100)	0 (0)	0 (0)	2 (100)
Proteus spp.	1 (50)	1 (50)	1 (50)	1 (50)	0 (0)	2 (100)
Proteus vulgaris	0 (0)	2 (100)	0 (0)	2 (100)	0 (0)	2 (100)
Providencia rettgeri	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Salmonella paratyphi	2 (66, 66)	1 (33, 33)	3 (100)	0 (0)	0 (0)	3 (100)
Salmonella paratyphi A	2 (100)	0 (0)	2 (100)	0 (0)	2 (75)	1 (25)
Salmonella spp.	5 (62, 5)	3 (37, 5)	8 (100)	0 (0)	1 (12, 5)	7 (87, 5)
Salmonella Typhi	2 (100)	0 (0)	2 (100)	0 (0)	0 (0)	2 (100)
Serratia liquefaciens	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Serratia spp.	0 (0)	2 (100)	0 ()	2 (100)	0 (0)	2 (100)
Shigella spp.	1 (50)	1 (50)	2 (100)	0 (0)	0 (0)	2 (100)
Yersinia enterocolitica	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)
Yersinia spp.	0 (0)	1 (100)	0 (0)	1 (100)	0 (0)	1 (100)

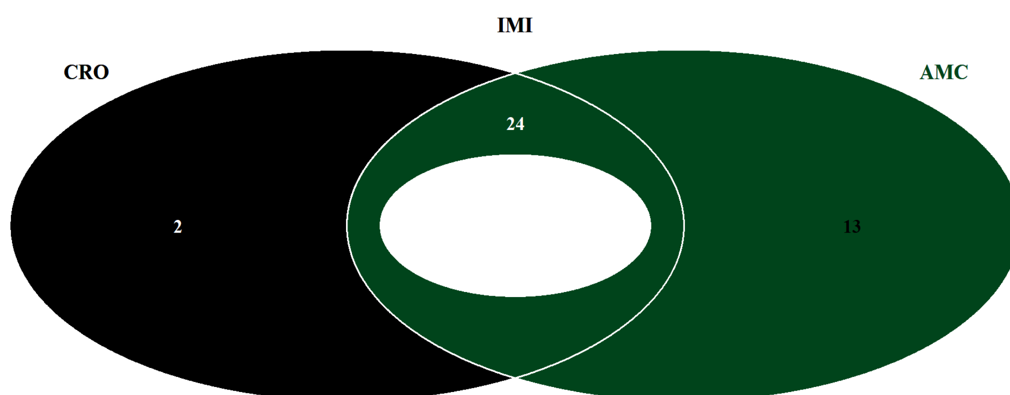

Figure 3. Distribution of the number of bacterial species according to their resistance to the three families of antibiotics.

3.2. Resistance Profile of Bacteria to the Three Families of Antibiotics According to Bat Species

When evaluating the bacterial resistance profile of bat species to Ceftriaxone, the presence of variability within bat species ($p < 0.05$) was found. Indeed, the species *R. alcyone*, *M. pumilus* and *H. jonesi* are the bat species that harboured bacteria that were all resistant to Ceftriaxone and therefore to third-generation cephalosporins (Table 4).

Table 4. Bacterial resistance profile of bat species in relation to Ceftriaxone.

Characteristic	R, N = 29[1]	S, N = 55[1]	Overall, N = 84[1]	p-value[2]
Bats				0.042
Epomophorus gambianus	11 (38%)	10 (18%)	21 (25%)	
Hyposideros caffer	2 (6.9%)	1 (1.8%)	3 (3.6%)	
Hyposideros jonesi	2 (6.9%)	0 (0%)	2 (2.4%)	
Mops condylurus	5 (17%)	16 (29%)	21 (25%)	
Mops midas	1 (3.4%)	14 (25%)	15 (18%)	
Mops pumilus	1 (3.4%)	0 (0%)	1 (1.2%)	
Pipistrellus nanulus	1 (3.4%)	2 (3.6%)	3 (3.6%)	
Rhinolophus alcyone	3 (10%)	0 (0%)	3 (3.6%)	
Scotophilus leucogaster	3 (10%)	12 (22%)	15 (18%)	

[1]n (%); [2]Kruskal-Wallis rank sum test.

Across all bat species, there was no difference in the resistance of their bacteria to amoxicillin plus clavulanic acid ($p > 0.05$). However, species such as *Hyposideros caffer*, *Hyposideros jonesi*, *Mops pumilus and Rhinolophus alcyone* harboured bacterial species that were all 100% resistant to amoxicillin and clavulanic acid (Table 5).

Table 5. Bacterial resistance profile of bat species in relation to amoxicillin and clavulanic acid.

Characteristic	R, N = 39[1]	S, N = 45[1]	Overall, N = 84[1]	p-value[2]
Bats				0.081
Epomophorus gambianus	12 (31%)	9 (20%)	21 (25%)	
Hyposideros caffer	3 (7.7%)	0 (0%)	3 (3.6%)	
Hyposideros jonesi	2 (5.1%)	0 (0%)	2 (2.4%)	
Mops condylurus	9 (23%)	12 (27%)	21 (25%)	
Mops midas	3 (7.7%)	12 (27%)	15 (18%)	
Mops pumilus	1 (2.6%)	0 (0%)	1 (1.2%)	
Pipistrellus nanulus	1 (2.6%)	2 (4.4%)	3 (3.6%)	
Rhinolophus alcyone	3 (7.7%)	0 (0%)	3 (3.6%)	
Scotophilus leucogaster	5 (13%)	10 (22%)	15 (18%)	

[1]n (%); [2]Kruskal-Wallis rank sum test.

As for imipenem, the leading carbapenem, there was no variability between bat species in terms of resistance. However, it appears that only the bacterial species isolated from *Mops condylurus* showed resistance to this family of antibiotics (Table 6).

Table 6. Bacterial resistance profile of bat species according to Imipenem.

Characteristic	R, N = 2[1]	S, N = 82[1]	Overall, N = 84[1]	p-value[2]
Bats				0.7
Epomophorus gambianus	0 (0%)	21 (26%)	21 (25%)	
Hyposideros caffer	0 (0%)	3 (3.7%)	3 (3.6%)	
Hyposideros jonesi	0 (0%)	2 (2.4%)	2 (2.4%)	
Mops condylurus	2 (100%)	19 (23%)	21 (25%)	
Mops midas	0 (0%)	15 (18%)	15 (18%)	
Mops pumilus	0 (0%)	1 (1.2%)	1 (1.2%)	
Pipistrellus nanulus	0 (0%)	3 (3.7%)	3 (3.6%)	
Rhinolophus alcyone	0 (0%)	3 (3.7%)	3 (3.6%)	
Scotophilus leucogaster	0 (0%)	15 (18%)	15 (18%)	

[1]n (%); [2]Kruskal-Wallis rank sum test.

3.3. Relationship between Bacterial Resistance to Antibiotics and the Sex of Their Hosts

The analyses show that the resistance of bacteria to the three families of antibiotics is not related to the sex of the bats. Indeed, the Pearson's Chi-squared test showed an absence of significant difference ($p > 0.05$). However, there was a strong tendency for bacteria isolated from female bats to be resistant to CRO (CRO residues = 1.12) and IMI (IMI residues = 0.13), whereas bacteria isolated from males showed a strong tendency for resistance to AMC (AMC residues = 1.38) (Figure 4).

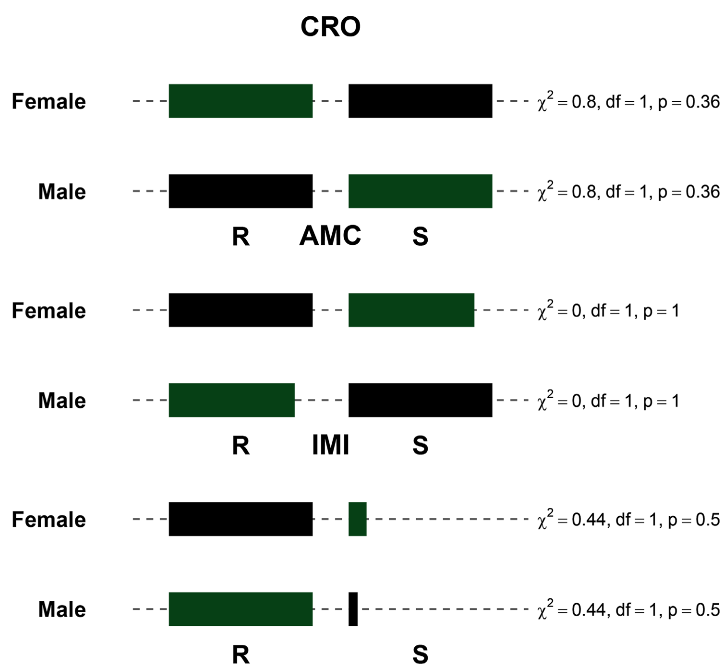

Figure 4. Residues and number of observations of resistance to the three families of antibiotics of isolated bacteria according to the sex of the bats with Pearson's Chi-squared test.

Among the 30 specimens of ROC-resistant bacteria, 43% and 57% were from female and male bats, respectively. Of the 40 specimens of AMC-resistant bacteria, 52% and 48% were from female and male bats, respectively. The two IMI-resistant bacteria were only isolated from female bats (Figure 5).

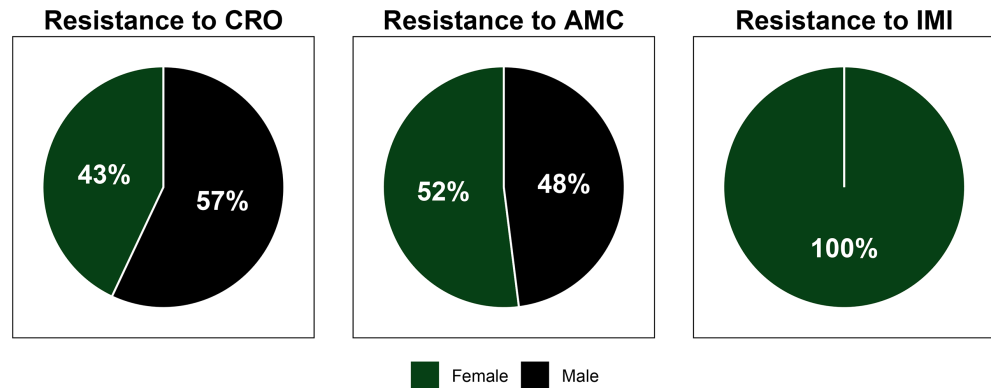

Figure 5. Percentage of bacterial resistance to the three families of antibiotics according to the sex of the bats.

Figure 6 shows the percentage of metallo-betalactamase (NDM) and Imipenemase (IMP) resistance genes in the population of bacterial strains. The NDM gene is abundant, with a rate of 97%, compared with 3% for the IMP gene.

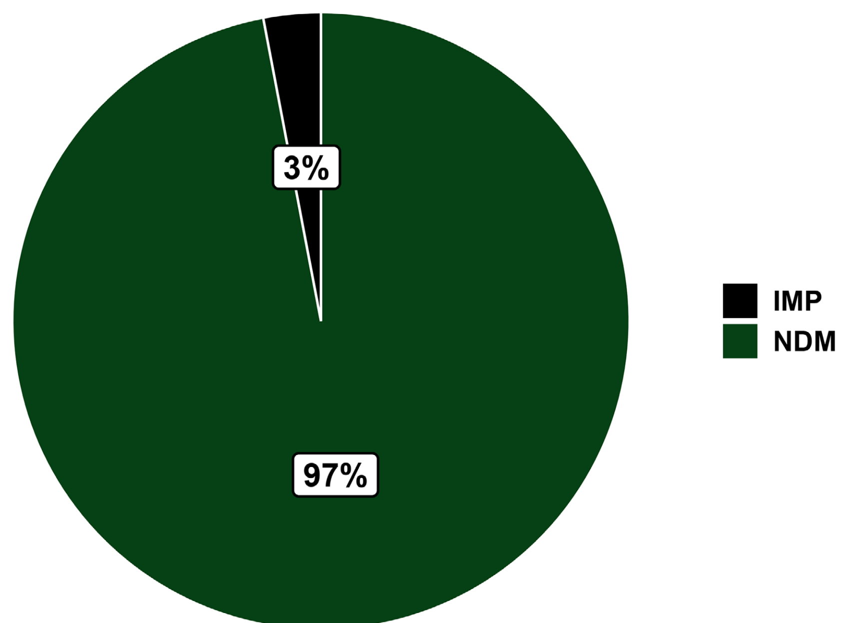

Figure 6. Percentage of resistance genes according to their occurrence.

4. Discussion

This survey confirms the bacterial richness of bats in Africa. Indeed, 29 species of

enterobacteria were isolated from 84 bat specimens. This confirms the results of several authors [15] [28] [29]. Enterobacteriaceae closely related to humans were isolated in this study, such as Salmonella, Shigella, and Yersinia. These bacteria are known to be responsible for several public health problems [28] [30] [31]. The presence of such bacterial richness in bats could be explained by their cohabitation with humans and also with wild birds which are known to be very rich in bacteria [32] [33]. The Enterobacteriaceae isolated in this study showed variability in resistance to the presence of the three families of antibiotics. The rate of resistance to CRO, AMC and IMI were 35% and 46%, respectively and 2.4%. Thus, this resistance of enterobacteria could be due to several factors. Indeed, the cohabitation between bats, wild birds and humans is a factor that favours resistance to antibiotics. [34] has shown that animals harbouring antibiotic-resistant Enterobacteriaceae are more frequently found in animals from the most anthropised sites. Various studies have shown the role of wild birds as a reservoir of antibiotic resistance. For example, migratory animals could contribute to the long-distance spread of antibiotic resistance in bats [15]. Recent findings suggest that wild animals may play a role in the origin of antibiotic resistance [15] [35] [36].

Moreover, even insects that are part of the diet of insectivorous bats may have a role in the bacterial resistance of bats through the presence of resistance genes in their microbiota according to [15].

We found low resistance of bacteria to IMI compared to the other two antibiotics. This could be explained by the absence of a resistance gene developed for this family of antibiotics.

Imipenem, a member of the broad-spectrum Carbapenem family, is known to be highly effective against bacteria [37]. Furthermore, it is difficult to implicate the nature of the medium in this explanation because the same bacteria isolated from bats in the same medium showed resistance to CRO and AMC.

The percentage of *E. coli* resistance was highest with all three antibiotics. Our results are similar to those of [38] who found that the genus Pteropus carried *E. coli* resistant to amoxicillin, carbapenems (*bla*NDM-5) and fluoroquinolone was detected in one E. coli isolate, and two isolates were resistant to third generation cephalosporins (*bla*CTX-M-27etampC).

Two species of bacteria, *Salmonella* spp. and *Salmonella paratyphi* A. were resistant to all three families of antibiotics. This resistance of the two salmonella species could be due to the pollution of the bats' living environments and the development of resistance genes. Four species of this genus were isolated during this study. This genus, in addition to having a high capacity for resistance, is very present in bats and wildlife. Although bacterial resistance to antibiotics may be caused by anthropogenic pollution, However, heavy metals or biocides can also contribute to the selection of bacterial resistance [6]. The Kruskal Wallis test showed that the resistance of the bacteria is not related to the host species of the

bats that harbour them. Indeed, bats live in colonies and often share the same biotopes.

Bacterial resistance was highest in bacteria isolated from *Epomophorus gambianus* species. This resistance was present in all three families of antibiotics. This result can be explained by the promiscuity of this bat species with humans. Indeed, in addition to being the most abundant bat species in Burkina Faso, it has a wide distribution [20]. Because of its frugivorous diet, it is present on trees such as mango and fig trees in search of fruit. The same trees are heavily exploited by humans so this could lead to transfer of resistance genes. Indeed, anthropogenic inputs into the environment can serve as sources of antimicrobial resistant bacteria and alter the ecology and population dynamics of wild animals [38]. Pearson's Chi-squared test showed that the sex of bats is not a parameter that influences the resistance of bacteria to antibiotics. Of course, male and female bats live together and reproduce, so they share the same bacteria and therefore the same resistance genes. However, it has been found that bacteria isolated from female bats have an easy resistance to AMC and IMI. Although male and female bats share the same ecological niches, females possess hormones that help them to be more resistant to the presence of pathogens. Thus, these same hormones could contribute to the birth of resistance genes.

The phenotypic expressions in relation to antibiotics led us to verify the presence or absence of two resistance genes responsible for the production of beta-lactamase enzymes: NDM (New Delhi Metallo-beta-lactamase) and IMP (imipenimase). Our results show that the NDM gene is highly present (97 %) in all strains, and is predominantly dominated by *E. coli*, unlike the *IMP* gene (3 %). Our results are similar to those of [7] Benavides *et al*., (2022) who found a high genetic distribution of betalactamases in bat populations. This similarity could be explained by the fact that pathogens are transmitted and conserved between wild animals, especially bats, due to their gregarious nature.

5. Conclusion

From this survey, which is the first in Burkina Faso, it was found that bats in Burkina Faso harbour enterobacteria with variable resistance patterns depending on the bat species and antibiotics. Of the 29 species of enterobacteria isolated from bats, *Salmonella typhi* and *Salmonella paratyphi* A showed resistance to all three antibiotics. The sex of the bats does not seem to be a factor influencing the resistance of their bacteria to antibiotics. Our results reinforce the need to monitor antimicrobial resistance in wildlife, in their environment and also to study the anthropogenic actions on this wildlife.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

[1] World Health Organization (2019) Antibacterial Agents in Clinical Development: An Analysis of the Antibacterial Clinical Development Pipeline. WHO.

[2] Medina-Pizzali, M.L., Venkatesh, A., Riveros, M., Cuicapuza, D., Salmon-Mulanovich, G., Mäusezahl, D., *et al.* (2022) Whole-Genome Characterisation of ESBL-Producing *E. coli* Isolated from Drinking Water and Dog Faeces from Rural Andean Households in Peru. *Antibiotics*, **11**, Article 692. https://doi.org/10.3390/antibiotics11050692

[3] Laborda, P., Sanz-García, F., Ochoa-Sánchez, L.E., Gil-Gil, T., Hernando-Amado, S. and Martínez, J.L. (2022) Wildlife and Antibiotic Resistance. *Frontiers in Cellular and Infection Microbiology*, **12**, Article 873989. https://doi.org/10.3389/fcimb.2022.873989

[4] Iovine, R.D.O., Dejuste, C., Miranda, F., Filoni, C., Bueno, M.G. and Carvalho, V.M.D. (2015) Isolation of *Escherichia coli* and *Salmonella* Spp. from Free-Ranging Wild Animals. *Brazilian Journal of Microbiology*, **46**, 1257-1263. https://doi.org/10.1590/s1517-838246420140843

[5] Rabinowitz, P., Scotch, M. and Conti, L. (2010) Human and Animal Sentinels for Shared Health Risks. *Veterinaria Italiana*, **45**, 23-24.

[6] Martínez, J.L. (2008) Antibiotics and Antibiotic Resistance Genes in Natural Environments. *Science*, **321**, 365-367. https://doi.org/10.1126/science.1159483

[7] Benavides, J.A., Godreuil, S., Opazo-Capurro, A., Mahamat, O.O., Falcon, N., Oravcova, K., *et al.* (2022) Long-Term Maintenance of Multidrug-Resistant *Escherichia coli* Carried by Vampire Bats and Shared with Livestock in Peru. *Science of The Total Environment*, **810**, Article ID: 152045. https://doi.org/10.1016/j.scitotenv.2021.152045

[8] Mühldorfer, K., Schwarz, S., Fickel, J., Wibbelt, G. and Speck, S. (2011) Genetic Diversity of Pasteurella Species Isolated from European Vespertilionid Bats. *Veterinary Microbiology*, **149**, 163-171. https://doi.org/10.1016/j.vetmic.2010.10.002

[9] Schiffmann, C., Unterhitzenberger, G. and Ortmann, S. (2020) Extrahepatic Biliary Tract Pathologies in Mammalian Species of Zoo Animals and Wildlife: A Review. *The Journal of Basic and Applied Zoology*, **81**, Article No. 20. https://doi.org/10.1186/s41936-020-00162-2

[10] Logan, L.K. and Weinstein, R.A. (2017) The Epidemiology of Carbapenem-Resistant Enterobacteriaceae: The Impact and Evolution of a Global Menace. *The Journal of Infectious Diseases*, **215**, S28-S36. https://doi.org/10.1093/infdis/jiw282

[11] Liu, M., Liu, J., Ma, J., Li, W., Zhao, X., Jia, W., *et al.* (2022) Antimicrobial Resistance and Molecular Characterization of Gene Cassettes from Class 1 Integrons in Carbapenem-Resistant *Escherichia coli* Strains. *Microbial Pathogenesis*, **170**, Article ID: 105669. https://doi.org/10.1016/j.micpath.2022.105669

[12] Beirão, B.C.B., Ingberman, M., Mesa, D., Salles, G.B.C., Muniz, E.C. and Caron, L.F. (2021) Effects of aroA deleted *E. coli* vaccine on intestinal microbiota and mucosal immunity. *Comparative Immunology, Microbiology and Infectious Diseases*, **75**, Article ID: 101612. https://doi.org/10.1016/j.cimid.2021.101612

[13] Jarlier, V., Nicolas, M.H., Fournier, G. and Philippon, A. (1988) Extended Broad-Spectrum β-Lactamases Conferring Transferable Resistance to Newer β-Lactam Agents in Enterobacteriaceae: Hospital Prevalence and Susceptibility Patterns. *Clinical Infectious Diseases*, **10**, 867-878.

https://doi.org/10.1093/clinids/10.4.867

[14] Ranjan, R. and Thatikonda, S. (2021) β-Lactam Resistance Gene NDM-1 in the Aquatic Environment: A Review. *Current Microbiology*, **78**, 3634-3643. https://doi.org/10.1007/s00284-021-02630-6

[15] Garcês, A. (2022) Bats and Antibiotic Resistance: A Culprit or a Victim? *World's Veterinary Journal*, **12**, 221-229. https://doi.org/10.54203/scil.2022.wvj28

[16] Garcês, A., Correia, S., Amorim, F., Pereira, J.E., Igrejas, G. and Poeta, P. (2019) First Report on Extended-Spectrum β-Lactamase (ESBL) Producing *Escherichia coli* from European Free-Tailed Bats (*Tadarida teniotis*) in Portugal: A One-Health Approach of a Hidden Contamination Problem. *Journal of Hazardous Materials*, **370**, 219-224. https://doi.org/10.1016/j.jhazmat.2017.12.053

[17] Garcês, A., Correia, S., Silva, V., Pereira, J.E., Amorim, F., Igrejas, G., *et al.* (2020) Detection of Antimicrobial Resistance in Faecal *Escherichia coli* from European Free-Tailed Bats (*Tadarida teniotis*) in Portugal. *Acta Chiropterologica*, **21**, 403-409. https://doi.org/10.3161/15081109acc2019.21.2.015

[18] Mbehang Nguema, P.P., Onanga, R., Ndong Atome, G.R., Obague Mbeang, J.C., Mabika Mabika, A., Yaro, M., *et al.* (2020) Characterization of ESBL-Producing Enterobacteria from Fruit Bats in an Unprotected Area of Makokou, Gabon. *Microorganisms*, **8**, Article 138. https://doi.org/10.3390/microorganisms8010138

[19] Obodoechi, L.O., Carvalho, I., Chenouf, N.S., Martínez-Álvarez, S., Sadi, M., Nwanta, J.A., *et al.* (2021) Antimicrobial Resistance in *Escherichia coli* Isolates from Frugivorous (*Eidolon helvum*) and Insectivorous (*Nycteris hispida*) Bats in Southeast Nigeria, with Detection of CTX-M-15 Producing Isolates. *Comparative Immunology, Microbiology and Infectious Diseases*, **75**, Article ID: 101613. https://doi.org/10.1016/j.cimid.2021.101613

[20] Thiombiano, N.G., *et al.* (2021) Bats Diversity and Abundance, Record of *Taphozous mauritianus* E. Geoffroy St.-Hilaire, 1818 for the First Time in Burkina Faso. *Journal of Entomology and Zoology Studies*, **9**, 26-31.

[21] Thiombiano, N.G., *et al.* (2020) Inventory of Bat (*Scotophilus leucogaster*, Cretzschmar, 1826) Ectoparasites of Savannah Area in Burkina Faso. *Journal of Biodiversity and Environmental Sciences*, **16**, 17-24.

[22] Miller, C. (2024) Host Specificity and Ectoparasite Load of Bat Flies in Utila, Honduras. University of New Orleans. https://scholarworks.uno.edu/honors_theses/63/

[23] Bergmans, W. (1997) Taxonomy and Biogeography of African Fruit Bats (Mammalia, Megachiroptera). 5. The Genera Lissonycteris, Myonycteris and Megaloglossus; General Remarks and Conclusions. *Beaufortia*, **47**, 11-90. https://repository.naturalis.nl/pub/504952

[24] Hayman, R.W. and Hill, J.E. (1971) Order Chiroptera. In: Meester, J. and Setzer, H.W., Eds., *The Mammals of Africa, an Identification Manual*, Smithsonian Institution, 1-73.

[25] Rosevear, D.R. (1965) The Bats of West Africa. Part 3: Microchiroptera. Trustees of the British Museum (Natural History).

[26] Drieux, L., Brossier, F., Sougakoff, W. and Jarlier, V. (2008) Phenotypic Detection of Extended-Spectrum β-Lactamase Production in Enterobacteriaceae: Review and Bench Guide. *Clinical Microbiology and Infection*, **14**, 90-103.

https://doi.org/10.1111/j.1469-0691.2007.01846.x

[27] Huang, S., Dai, W., Sun, S., Zhang, X. and Zhang, L. (2012) Prevalence of Plasmid-Mediated Quinolone Resistance and Aminoglycoside Resistance Determinants among Carbapeneme Non-Susceptible *Enterobacter cloacae*. *PLOS ONE*, **7**, e47636. https://doi.org/10.1371/journal.pone.0047636

[28] Mühldorfer, K. (2012) Bats and Bacterial Pathogens: A Review. *Zoonoses and Public Health*, **60**, 93-103. https://doi.org/10.1111/j.1863-2378.2012.01536.x

[29] Nowak, K., Fahr, J., Weber, N., Lübke-Becker, A., Semmler, T., Weiss, S., *et al.* (2017) Highly Diverse and Antimicrobial Susceptible *Escherichia coli* Display a Naïve Bacterial Population in Fruit Bats from the Republic of Congo. *PLOS ONE*, **12**, e0178146. https://doi.org/10.1371/journal.pone.0178146

[30] Mühldorfer, K., Speck, S. and Wibbelt, G. (2014) Proposal of *Vespertiliibacter pulmonis* Gen. Nov., Sp. Nov. and Two Genomospecies as New Members of the Family *Pasteurella*ceae Isolated from European Bats. *International Journal of Systematic and Evolutionary Microbiology*, **64**, 2424-2430. https://doi.org/10.1099/ijs.0.062786-0

[31] Doutre, M.P. and Sarrat, H. (1973) Sérotypes de Salmonelles isolées chez les chiroptères frugivores et insectivores du Sénégal. Importance épidémiologique. *Revue d'élevage et de médecine vétérinaire des pays tropicaux*, **26**, 279-287. https://doi.org/10.19182/remvt.7835

[32] Oteo, J., Diestra, K., Juan, C., Bautista, V., Novais, Â., Pérez-Vázquez, M., *et al.* (2010) Corrigendum to "Extended-Spectrum *β*-Lactamase-Producing *Escherichia coli* in Spain Belong to a Large Variety of Multilocus Sequence Typing Types, Including ST10 Complex/a, ST23 Complex/A and ST131/B2" [Int. J. Antimicrob. Agents 34 (2009) 173-176]. *International Journal of Antimicrobial Agents*, **36**, 483. https://doi.org/10.1016/j.ijantimicag.2010.07.001

[33] Stępień-Pyśniak, D., Hauschild, T., Kosikowska, U., Dec, M. and Urban-Chmiel, R. (2019) Biofilm Formation Capacity and Presence of Virulence Factors among Commensal *Enterococcus* spp. from Wild Birds. *Scientific Reports*, **9**, Article No. 11204. https://doi.org/10.1038/s41598-019-47602-w

[34] Di Lallo, G., D'Andrea, M.M., Sennati, S., Thaller, M.C., Migliore, L. and Gentile, G. (2021) Evidence of Another Anthropic Impact on *Iguana delicatissima* from the Lesser Antilles: The Presence of Antibiotic Resistant Enterobacteria. *Antibiotics*, **10**, Article 885. https://doi.org/10.3390/antibiotics10080885

[35] Ramasindrazana, B., Dellagi, K., Lagadec, E., Randrianarivelojosia, M., Goodman, S.M. and Tortosa, P. (2016) Diversity, Host Specialization, and Geographic Structure of Filarial Nematodes Infecting Malagasy Bats. *PLOS ONE*, **11**, e0145709. https://doi.org/10.1371/journal.pone.0145709

[36] Fiona K. McDougall, M.L.P. and Boardman, W.S.J. (2021) Characterization of Beta-lactam-Resistant *Escherichia coli* from Australian Fruit Bats Indicates Anthropogenic Origins. *Microbial Genomics*, **7**, 539-547. https://doi.org/10.1099/mgen.0.000571

[37] Batista, J.M.N. (2023) Detecção de *Leptospira* spp. e *Enterobacteriales* em morcegos (Chiroptera, Mammalia) da região urbana de São Paulo, SP, Brasil. Master's Thesis, Universidade de São Paulo. https://doi.org/10.11606/D.10.2023.tde-14112023-115052

[38] Ahlstrom, C.A., van Toor, M.L., Woksepp, H., Chandler, J.C., Reed, J.A., Reeves, A.B., *et*

al. (2021) Evidence for Continental-Scale Dispersal of Antimicrobial Resistant Bacteria by Landfill-Foraging Gulls. *Science of the Total Environment*, **764**, Article ID: 144551. https://doi.org/10.1016/j.scitotenv.2020.144551