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Let sleeping dogs lie? Domestic carnivores as potential sentinels for One Health surveillance of antimicrobial resistance: the example of Réunion Island

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Objectives: Extended-spectrum β-lactamase- and carbapenemase-producing Enterobacterales (ESBL-E and CP-E, respectively) are global public health threats, prioritized by the WHO for urgent action. We assessed prevalence, risk factors and genomic context of ESBL-E and CP-E carriage in domestic carnivores on Réunion Island and compared resistant isolates with human isolates to evaluate pets' role in islandwide antimicrobial resistance transmission.

Methods: Combining the methods of classical microbiology (selective culture, MS identification, disc-diffusion antimicrobial susceptibility testing) and WGS (using in-house Nanopore technology), we compared 55 ESBL-E isolates originating from 881 domestic carnivores obtained in an island-wide cross-sectional survey with 40 human isolates obtained through an existing ESBL surveillance network and from healthy volunteers. We evaluated the risk factors for ESBL-E carriage in domestic carnivores by fitting multivariate generalized linear models applied to data from anamnestic questionnaires.

Results: The prevalence of ESBL-E/CP-E carriage was 6% in carnivores (dogs: 8%; cats: 3%). The dominant ESBL gene across hosts (dogs, cats, humans) was $bla_{\text{CTX-M-15}}$, followed by $bla_{\text{CTX-M-27}}$. Three carbapenem-resistant isolates were detected in dogs, including the $bla_{\text{NDM-1}}$ gene. Risk factors for animal ESBL-E/CP-E colonization included intact reproductive status, recent antibiotic therapy and free-roaming behaviour. Animal-originating ESBL-producing *Escherichia coli* exhibited greater genetic diversity than human-origin isolates, suggesting multiple introduction sources and environmental influences.

Conclusions: Our findings suggest that domestic carnivores should be routinely included in surveillance networks, as their lifestyle and close contact with humans may allow trends in their enteric antimicrobial-resistant flora to foreshadow trends in the human resistome.

Introduction

Antimicrobial resistance (AMR) can be transmitted from animals to humans via direct contact or shared environments. Through horizontal gene transfer, the resistance determinants carried by commensal bacteria can be acquired by pathogens, undermining treatment effectiveness. To better understand these dynamics and curb further spread, resistant bacteria must be

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monitored at the molecular level by integrated One Health surveillance systems, enabling real-time comparisons of AMR trends across different epidemiological compartments.[1–4] ESBL- and carbapenemase-producing Enterobacterales (ESBL-E and CP-E) are classified by the WHO as critical priority pathogens for which new antibiotics are urgently needed.[5] In line with this, WHO's 'Tricycle protocol' recommends the ESBL-producing *Escherichia coli* as a suitable indicator for tracking AMR spread in humans, animals and the environment.[6]

AMR surveillance in domestic carnivores is often overlooked, yet these animals serve as crucial epidemiological links between humans, livestock and wildlife due to their close interactions with all three.[7,8] A stray dog, for example, might spend years scavenging in dumpsters before being adopted and finding its way into the bed of his owner. Similarly, a cat lounging on a kitchen counter could have spent the night interacting with wildlife; encounters of the kind that its owner would never experience. Such transitions and interfaces are not rare on Réunion Island, a melting pot of Indian, East African and European communities, trade routes and cultures (including microbial cultures).[9]

In our study, we investigated ESBL-E and/or CP-E carriage in domestic carnivores on Réunion Island, considering two plausible hypotheses for colonization: selective pressure from direct antibiotic therapy (given the island's advanced veterinary care) and passive colonization from exposure to human- or livestock-contaminated environments. We analysed faecal swabs collected through a network of veterinary clinics and animal protection organizations, comparing the genetic profiles of ESBL-E and CP-E isolates with isolates of human origin from outpatient facilities in the same area. We discuss the observed overlaps and their potential epidemiological implications.

Materials and methods

Study area and methodological workflow

Réunion is a French overseas territory in the south-west Indian Ocean region. This small (>2500 km^2 and close to 900 000 in population) but highly connected tropical island faces ongoing challenges due to a substantial stray and free-roaming dog and cat population, with a dog-to-human ratio estimated as 1:4.[10]

In the spirit of One Health collaboration, the analysis of multiresistant strains of both animal and human isolates was performed by a single multidisciplinary research team, using identical methodologies and laboratory benches (Figure S1, available as Supplementary data at *JAC* Online).

Animal sampling

The collection of animal samples was conducted between January and November 2023 in partnership with 17 veterinary clinics and two animal protection NGOs (Table S1, Figure S2), all invited to participate in the research project on a voluntary basis. All collaborators were instructed to collect rectal swabs from dogs and cats, regardless of their health status, age, gender or ownership. Each animal was sampled once, and hospitalized animals were sampled on admission. Animal owners or keepers signed a written informed consent before sampling. All samples were collected either from freshly deposited faeces, or as part of routine screening during clinical interventions when animals were anaesthetized for the purpose of sterilization or other surgical procedures.

The swabs were immediately placed in Amies medium supplied by the manufacturer (Amies swab PS + Viscose, Deltalab, Spain) and refrigerated for a maximum of 1 week before analysis. Every swab was accompanied by an anamnestic questionnaire (Table S2), filled in by the veterinarian or veterinary assistant, either in Kobo Toolbox[11] online form or on paper (the latter transcribed to Kobo by an experienced data manager upon the sample reception), always in collaboration with animal owners. All variables evaluated as potential risk factors for the CP-E/ESBL-E carriage were extracted from this questionnaire (Supplementary data S1).

Human sampling

During the same period, 93 ESBL-E isolates were randomly selected from the regional network of diagnostic laboratories coordinated by the University Hospital of Réunion Island. This network represents >90% of the samples analysed in local diagnostic laboratories. Samples (swabs from respiratory, urogenital or digestive tracts, swabs from wounds) are taken for diagnostic purposes, and ESBL isolates, if obtained, are routinely archived to support the ESBL-surveillance network. Patients are always informed about the possibility of using the samples for research purposes under anonymity conditions; this study handled only archived bacterial isolates and no human subjects. For the purpose of anonymity, the anamnestic data pertaining to the isolates' origin were not handled in this study. Only outpatient-originating isolates were included in this population sample, to represent the situation in the community.

In addition to strains obtained in the frame of the surveillance network, six swabs were provided by healthy volunteer collaborators within the research group and analysed in the same manner as animal-originating samples (Figure S1).

Culture and isolate selection

Human-originating isolates were streaked on CHROMID ESBL from cryobeads; visual examination of individual colonies obtained by standard quadrant streak was performed to verify the culture purity before further analysis.

Animal swabs were streaked directly on the selective agars, CHROMID ESBL and CHROMID CARBA SMART (bioMérieux, France), and incubated at 37°C for 24 h under aerobic conditions. Purple, blue and turquoise colonies (as per the manufacturer's instructions for Enterobacterales) were isolated and approximately two loops of the pure culture were frozen at −80°C in 1 mL of glycerol-peptone-based cryoprotective medium for further analyses. The cryoprotective medium was prepared as follows: 1 g peptone (Gibco, USA), 0.5 g NaCl (Sigma-Aldrich, Germany), 25 mL glycerol ≥99% (Merck, Germany), and 75 mL distilled water, for a final volume of 100 mL.

White colonies morphologically similar to *E. coli* were tested for oxidase production (Oxidase Test Stick, Liofilchem, Italy) and frozen if negative. In cases where two or more morphologically different colonies (different shape or colour) were obtained from the same animal, every presumed strain was stored separately.

Bacterial species identification and resistance testing

The species identification was performed by MALDI-TOF MS (Microflex LT, Bruker Daltonics, Bremen, Germany), according to the manufacturer's instructions using the direct colony transfer method. Colony fragments from overnight cultures at 37°C were spotted onto the target, overlaid with 1 µL of HCCA-matrix (α-cyano-4-hydroxy-cinnamic acid; Bruker Daltonics) and air-dried. MS analysis was performed and the spectra obtained were compared with the database (MBT-IVD-Library-Revision J; Bruker) for species identification.

Only isolates of *E. coli*, *Klebsiella pneumoniae* and *Enterobacter* spp. were submitted for the subsequent molecular testing, because these taxa are the most frequently encountered Enterobacterales in clinical and surveillance isolates and are established sentinel species for carriage of ESBL and carbapenemase determinants.[3]

Antimicrobial susceptibility testing was performed by the disc diffusion method (SIRSCAN PREP, KIT CLSI/EUCAST; i2a, France), using a panel of 16 antibiotics (Table S3) on Mueller–Hinton plates (bioMérieux, France), according to EUCAST recommendations.[12] The 16 antibiotics were selected to find a maximum overlap between the recommendations of EARS Vet[3] and studies previously performed on La Réunion Island,[13] in order to allow for analytical continuity. The double-disc synergy test and combined disc test were performed as described previously,[14] to further specify the phenotypes.

Molecular analyses

We aimed to select up to 100 ESBL-E/CP-E isolates from the initial sample set for molecular analysis. ESBL-E isolates were chosen by resistance phenotype: for each host, we selected the isolates resistant to the largest number of antibiotic classes. All CP-E were subjected to molecular analysis. DNA was extracted from approximately 10 µL of overnight culture obtained on selective media (CHROMID-ESBL or CHROMID-CARBA, according to the phenotype), using QIAmp DNA Mini-Kit (QIAGEN, USA), according to the manufacturer's recommendations, with several modifications (three washes with PBS at the beginning of the protocol, lysis at 57°C for 1 h). DNA concentrations were quantified using the Qubit 4.0 instrument (Invitrogen, USA) with the dsDNA HS 1× kit, following the manufacturer's instructions. DNA was diluted to 20 ng/µL using nuclease-free water.

The first 11 ESBL-E isolates obtained were submitted for Illumina sequencing at Genesupport (Fasteris Lifescience Services, Switzerland; service DNA-Seq, mechanical shearing 350 bp, sequencing instrument Miseq, sequencing run 2 × 150, proportion of PhiX 1%, run yield in million cluster 20, post-processing demultiplexing). This sample subset was later sequenced in-house by MinIon Mk1C (Oxford Nanopore Technology, UK) with rapid barcoding (SQK-RBK114.94) following the manufacturer's instructions. Negative barcode controls were included to exclude the possibility of barcode cross-talk. In-house comparisons between Illumina- and Nanopore-only-generated genome assemblies showed that all 11 compared genomes had consensus qualities of between Q55 and Q60 (1 error in every 300 000 to 1 000 000 bp), suggesting that Nanopore-only consensus genomes were highly accurate in the context of this study (data not shown).

Raw Nanopore data were basecalled and demultiplexed using Guppy v6.5.7.[15] Filtlong[16] was used to exclude sequences shorter than 400 bp or below Q10. Negative controls included at every step of the analysis did not generate any data. Flye[17] was used for genome assembly, allele profiles were determined by mlst,[18] resistance genes were detected by AMR FinderPlus,[19] and plasmidic or chromosomal origin of the contigs was estimated by PlasForest.[20]

Statistical analysis

The database (Supplementary data S1) of potential risk factors for ESBL-E/CP-E carriage in animals was created by extracting the questionnaire data from Kobo and adding the results of microbiological analyses. Initial data cleaning (harmonization of spelling of local names) was performed manually in Excel; some variables were later re-categorized in R. The analysis was performed in R[21]/R Studio.[22] The response variable was defined as 'carriage of ESBL-E and/or CP-E', coded 0 and 1. Several explanatory variables were considered as potential risk factors (namely sex, reproductive status, age, recent antibiotic therapy, access to outdoors, region, contact with other animals; see Supplementary data S2).

Two binomial GLMs (generalized linear models, using the default logit link) were constructed for cats and dogs to account for differences in their lifestyles, in both cases excluding variable interactions to prevent overfitting. Missing data were included in the analysis by treating them as a distinct category of the risk factors to minimize data loss. Model validity was ensured by checking for overdispersion and visually inspecting the distribution of Pearson residuals. Model selection was guided by expert judgement in conjunction with statistical criteria and model fit evaluation (avoiding automated selection procedures, to integrate both objective statistical measures and contextual understanding).

The contribution of each explanatory variable to the model was examined using type II analysis of variance (ANOVA),[23] and for variables with more than two levels, pairwise comparisons were conducted using Tukey's method for adjusting P values.[24] These comparisons were performed on the log odds ratio scale. Results were interpreted based on effect sizes, CIs of the estimated marginal means, the 5% threshold for significance and their relevance to the research context.

Results

Study sample

In total, the study included 93 preselected ESBL-E isolates of human origin, 6 swabs of healthy human volunteers and 881 swabs of animal origin, comprising 522 dogs and 359 cats (Figure S1). Animals from the northern and western sectors of the island were represented by 53% and 37% of the total, respectively. As per the admission reason, 62% of dogs and 84% of cats were sampled while surgically sterilized. Phenotypical analyses of the swabs recovered revealed 43 ESBL-E from dogs (28 *E. coli*, 11 *K. pneumoniae* and 4 *Enterobacter* sp.), 12 ESBL-E from cats (6 *E. coli*, 3 *K. pneumoniae* and 3 *Enterobacter* sp.) and 2 ESBL-producing *E. coli* from healthy human volunteers. In addition, four CP-E were found: two *Enterobacter* spp. and one *E. coli* in dogs and one *E. coli* of human origin.

Genetic background of ESBL-producing isolates in humans and animals, and overlaps thereof

Ninety-five ESBL-E or CP-E of human and animal origin ($n=40$ and $n=55$, respectively) were subjected for molecular analysis. These were *E. coli* (35, 28 and 6 of human, dog and cat origin, respectively), *K. pneumoniae* (1, 11 and 3 of human, dog and cat origin, respectively) and *Enterobacter* spp. (4, 4 and 3 of human, dog and cat origin, respectively). This section features only some highlights of the resistance profiles; complete results can be consulted in Supplementary data S3.

Across the host spectrum and bacterial species, the most common ESBL gene was $bla_{\text{CTX-M-15}}$ (47 isolates; 33× on a chromosome and 14× on a plasmid), followed by $bla_{\text{CTX-M-27}}$ (19 isolates; 7× on a chromosome and 12× on a plasmid) (Figure 1).

A phylogenetic analysis of all ESBL-producing and/or carbapenem-resistant *E. coli* strains revealed two major branches, one with predominance of animal-originating strains and other with predominance of human-originating strains (Figure 2).

No clones shared across compartments or individuals were detected. However, among the *E. coli* isolates, seven ST-ESBL combinations, including ST131, ST3580, ST219, ST457, ST1193 and ST7401, were observed in both dog and human isolates; these combinations involved $bla_{\text{CTX-M-15}}$ and $bla_{\text{CTX-M-27}}$ (Figure S3). No similar overlap was observed between cats and humans.

Among the CP-E isolates, we identified $bla_{\text{IMI-4}}$ in one isolate (dog-originating *Enterobacter sichuanensis*) and $bla_{\text{NDM-1}}$ in three isolates (human-originating *E. coli* ST1588, dog-originating *E. coli*

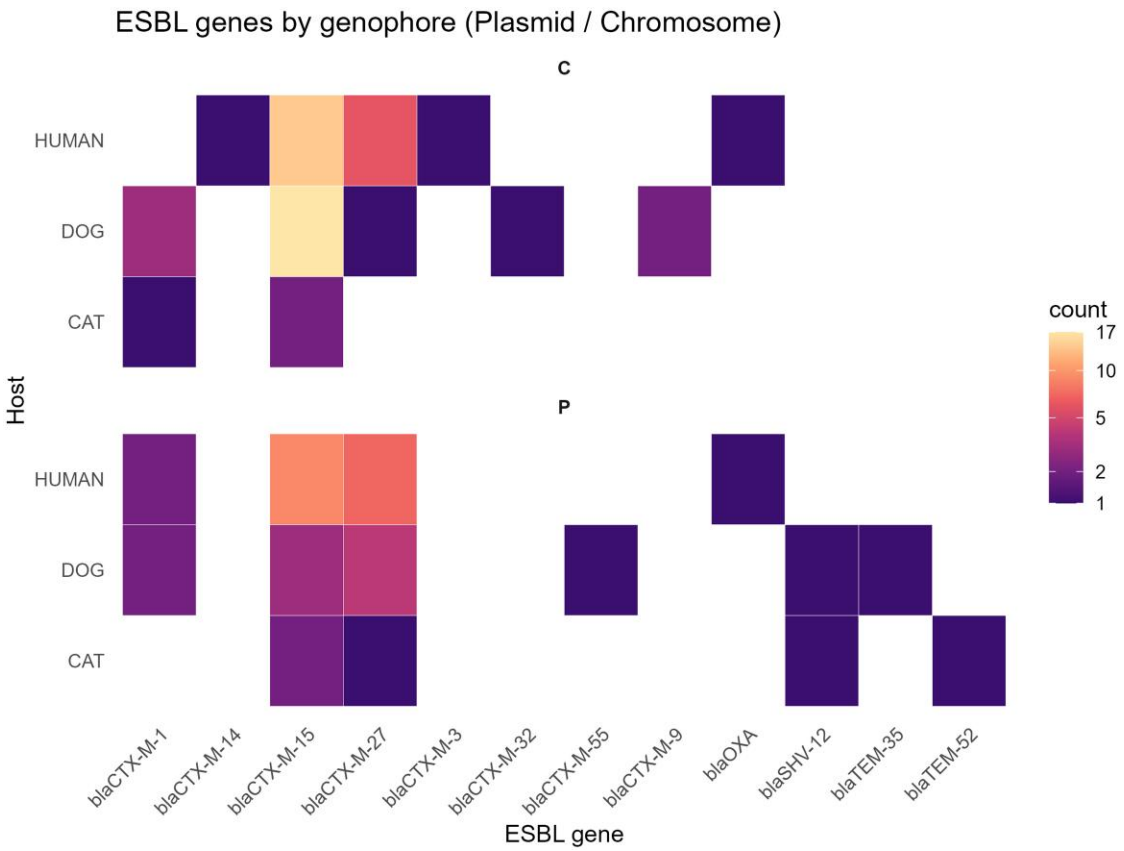


Figure 1. Heatmap showing the number of host-ESBL gene occurrences in Enterobacterales isolates (bacterial species not shown; data across isolates of *E. coli*, *K. pneumoniae* and *Enterobacter* spp.). Colour indicates $\log_{10}(1+n)$ (n = raw count); white tiles indicate no observed combinations. Panels show genophore: P = plasmid, C = chromosome. The figure illustrates that $bla_{\text{CTX-M-15}}$ and $bla_{\text{CTX-M27}}$ were the most frequent across all compartments of the dataset, both plasmid- and chromosome- mediated.

ST1193 and dog-originating *Enterobacter asburiae*). The latter two were carried by the same animal, which also harboured another ESBL-producing *E. coli* ST1193 (Supplementary data S3).

Prevalence and risk factors of ESBL-E/CP-E carriage in carnivores

The observed prevalence of carnivores colonized by ESBL-E/CP-E was 6% (95% CI, 4.65–7.83). It was relatively higher in dogs (8%; 95% CI, 5.9–10.6) than in cats (3%; 95% CI, 1.5–5.2).

The final model for the subset of dogs (Pearson residual-based dispersion parameter = 1.023) included the variables region, contact/cohabitation with other animals, recent antibiotic therapy, confinement or free access to outdoors, age and reproduction status. For cats, the final model (dispersion parameter = 0.69) included region, contact, antibiotic therapy, confinement and reproduction status.

In dogs, the factors associated with an increased probability of colonization by ESBL-E/CP-E were intact reproductive status, unconfined access to outdoors, and a history of antibiotic therapy within the past 6 months. For cats, lack of confinement was the only risk factor identified at the 0.05 *P*-value significance level (Table 1).

Discussion

Resistome overlaps across human and animal epidemiological compartments

Our study benefits from comparable genomic data derived from human and animal sources, thus allowing for meaningful parallels to be drawn. Although we did not recover any identical clones shared between humans and domestic carnivores, we observed notable overlaps in STs and/or resistance genes across compartments. Furthermore, we observed isolates with identical ST/ESBL combinations shared between dogs and humans. These overlaps are epidemiologically important, as they point to shared transmission networks and strongly suggest interspecies dissemination of resistance, reinforcing the need for integrated One Health surveillance.

Similarly to other studies,[25–27] $bla_{\text{CTX-M-15}}$ was by far the most prevalent ESBL gene, occurring both in humans and animals, followed by $bla_{\text{CTX-M-27}}$, $bla_{\text{CTX-M-1}}$ and $bla_{\text{CTX-M-14}}$, also present across the three studied compartments. Interestingly, these common ESBLs were often found on chromosomes, adding to the increasing number of reports portraying chromosomal integration of these originally plasmid-borne genes.[28]

E. coli STs most often detected in our domestic carnivores dataset (ST38, ST131, ST68, ST405, ST617 and ST648) are not

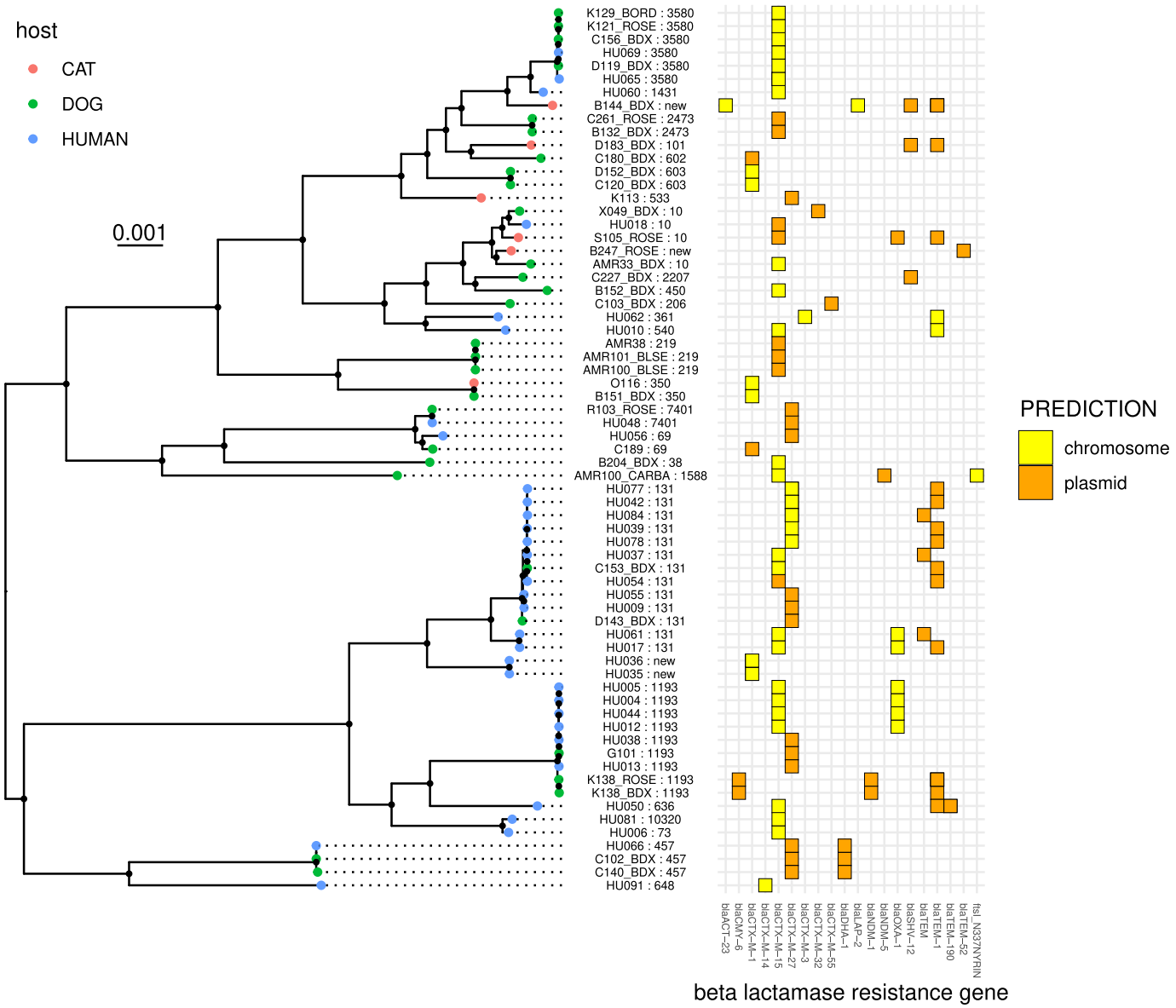


Figure 2. Whole genome SNP-based phylogeny of ESBL- and carbapenemase-producing *E. coli* isolates from this study. Isolate IDs and their associated MLST profiles are denoted. *Left:* Phylogenetic tree showing genetic similarity between isolates. Scale units are mutations per site. Tip colours denote host associations, following the legend. Note that fine-grain variation between members of the same ST is not necessarily apparent at this scale. *Right:* β-Lacatamase-resistance–associated genes. Colour indicates plasmid- or chromosomal-association as predicted by mobsuite.

Table 1. Overview of risk factors of ESBL-E/CAR-E carriage in domestic carnivores

Model	Variable	Contrast	OR [95% CI]	*P* value
Dogs	Reproduction status	Intact/neutered	8.16 [2, 33.3]	0.0034
	Confinement	No/yes	2.39 [1.06, 5.39]	0.0363
	Antibiotic therapy	No/yes	0.304 [0.01, 0.99]	0.0486
Cats	Confinement	No/yes	19.7 [1.51, 257]	0.0231

Interpretation aid: All else being equal, the model estimates that the odds of colonization by ESBL-E/CP-E are higher in intact dogs compared with neutered dogs and in non-confined dogs compared with confined dogs, whereas dogs without a history of antibiotic therapy within the previous 6 months have lower odds of colonization. These associations are statistically significant at the 5% level. In cats, lack of confinement is associated with higher odds of colonization by ESBL-E/CP-E, although the CI is wide; this association is statistically significant at the 5% level.

uncommon among pets' enteric microflora. On the other hand, ST1588, ST206, ST2473, ST450 and ST603 were rarely observed in carnivores previously.[27] We did not assess virulence factors in this study, but we note that ST131, ST38, ST405, ST617 and ST648 (all found in carnivores) are known to include lineages pathogenic to humans.[29]

Our phylogenetic assembly of ESBL-producing *E. coli* strains reveals two distinct clusters. The cluster containing predominantly human-origin strains shows (lower part of Figure 2) a higher degree of clonality, suggesting direct or limited-source contamination. In contrast, the cluster with the majority of animal-originating strains (upper part of Figure 2) displays greater genetic diversity, indicating that ESBL-producing *E. coli* transmission in companion animals is likely not clonal; instead, it may result from multiple introductions from sources like livestock and humans, compounded by selective pressures from veterinary antibiotic use.

As per the carbapenem resistance–conferring genes, while $bla_{\text{NDM-1}}$ is not entirely new among domestic carnivores,[30] our study seems to be the first report of the $bla_{\text{IMI-4}}$ class A carbapenemase-encoding gene occurrence therein.

ESBL carriage prevalence in domestic carnivores and associated risk factors

The prevalence of ESBL-E/CP-E observed in our study aligns with the 'global prevalence' reported for dogs and cats in a recent meta-analysis.[27] It is rather similar to European contexts[31,32] than to tropical countries. Our findings contrast with a previous study conducted in Réunion Island during 2019–2020,[13] where the observed prevalence was much lower (0.6% in dogs and 1.4% in cats). This could be due to an objective increase in ESBL-E prevalence such as observed in other studies,[32,33] or due to the inclusion of stray animals in our population sample.

While some encouraging reports show a decrease in ESBL prevalence following improved antibiotic stewardship in livestock in certain regions, including Réunion,[34–36] achieving similar reductions in companion animals might be a much more complex challenge. Unlike livestock, which are managed under controlled conditions with limited movement, the gut microbiota of free-roaming domestic carnivores is influenced by a diverse array of factors beyond antibiotic exposure alone, such as environmental sources and close contact with humans. It is plausible that environmental sources are especially important in humid, tropical climates that favour bacterial survival in the environment.

For dogs, and to some extent even for cats in our study, free outdoor access or a history of stray status suggested an increased risk of ESBL-E/CP-E colonization, likely due to frequent encounters with environmental reservoirs, such as soil, water and waste. Supporting these findings, Marchetti *et al.*[37] previously observed higher resistance rates in stray dogs compared with owned dogs. AMR thus adds another reason for management of the domestic carnivore population; promoting and facilitating neutering/spaying can help prevent increases in stray populations and reduce the risk of AMR transmission.

Regarding reproductive status, intact dogs were found to be at higher risk, likely due to hormone-driven behaviours, such as roaming and interaction with other animals. Finally, and not surprisingly, recent antibiotic treatment was a significant risk factor in dogs. It is also worth noting that a history of stray status may correlate with antibiotic exposure, as stray animals are more frequently exposed to infectious diseases (e.g. ehrlichiosis) that require antibiotic treatment before the animal rehoming. Such a combination of prior antibiotic therapy and stray history was observed in previous studies.[38]

Study limitations

Despite a considerable sample size, our study faced some important limitations. Firstly, we considered our carnivore population sample as representative of the island-wide population in terms of sex, age class, and confinement level, with proportions aligning closely to previous observations.[10,39,40] However, it was not representative from the geographical point of view (over-representation of the northern and western regions of the island) and hence the observed prevalence shall not be extrapolated for the whole of Réunion Island.

Secondly, while our statistical approach to evaluate the risk factors of ESBL-E/CP-E carriage provides some valuable insights, these should be interpreted with caution, due to the exclusion of interaction terms between the variables analysed, potential unmeasured confounding factors and, in some cases, the reliance on owners' self-reported information about previous antibiotic treatments in their animals. While the performance of the dog model seemed satisfactory, the cat model should be interpreted with caution, considering the wide CIs and relatively low dispersion parameter, indicating lack of data. Lastly, due to scarcity of CP-E isolates, we confounded ESBL-E and CP-E into one response variable, which could have masked some differences between these resistance profiles.

Are domestic carnivores sentinels and/or reservoirs of AMR?

The frequent usage of 'reservoir' or 'sentinel' when relating to the role of different animals in the epidemiological surveillance of AMR has been criticized for lacking sufficient evidence to meet the epidemiological definitions.[27] Our study's comparison of the genetic backgrounds of isolates from animals and humans —particularly the greater phylogenetic diversity observed in animal-originating isolates—strongly suggests that domestic carnivores on Réunion Island could indeed play the role of reservoir or sentinel species in AMR.

However, to establish true proof of such a concept and to leverage domestic carnivores as indicators of AMR trends, longitudinal and methodologically consistent data are needed. By tracking temporal trends across the three compartments—humans, animals and the environment—we can analyse potential synchronicity in resistome patterns and infer the directionality of intercompartmental transmissions. Such monitoring should routinely include free-roaming domestic carnivores to enable further investigation of our proposed hypothesis that these animals may become passively contaminated and subsequently disseminate antimicrobial-resistant microflora.

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Transparency declarations

None to declare.

Author contributions

Clément Rabaly (Conceptualization, Data curation, Formal analysis, Investigation, Methodology), David A. Wilkinson (Conceptualization, Data curation, Formal analysis, Methodology, Software, Supervision), Guillaume Miltgen (Conceptualization, Formal analysis, Writing—review & editing), Olivier Belmonte (Conceptualization, Resources), Anaïs Ethèves (Conceptualization, Investigation, Methodology, Project administration, Supervision, Writing—review & editing), Cléa Maurines-Carboneill (Investigation, Methodology), Axelle Sellier (Formal analysis, Investigation), Anna Doizy (Data curation, Formal analysis, Writing—review & editing), Adrien Rieux (Conceptualization, Methodology), Thomas Garrigos (Formal analysis), Cécile Squarzoni (Conceptualization) and Katerina Albrechtova (Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Project administration, Writing—original draft).

Supplementary data

Figures S1 to S3, Tables S1 to S3, and Supplementary data S1 to S3 are available as Supplementary data at *JAC* Online.

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