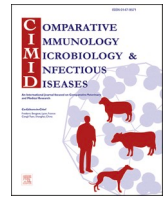




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Active surveillance of antimicrobial resistance in companion animals: A pilot study in a Spanish Veterinary Teaching Hospital

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ABSTRACT

The role of small animal veterinary hospitals in the onset and dissemination of antimicrobial-resistant organisms (AMROs) is still not clear, and the implementation of an internal surveillance systems is a cost-effective tool to better understand their impact. The aim of this study was to describe a pilot program of active surveillance in a Spanish Veterinary Teaching Hospital, developed to estimate the detection frequency of AMROs in the commensal flora of patients and in the environment. Surveillance was focused on Methicillin-resistant Staphylococci (MRS), third generation cephalosporins resistant gram-negative bacteria (3GCR-GNB), and carbapenems-resistant gram-negative bacteria (CR-GNB). Oral and perirectal swabs were collected in the same dogs and cats hospitalized > 48 h, at their admission and before their discharge. Out of 50 patients sampled, 24% (12/50) were carriers at admission of at least one of the three investigated AMROs. Twenty-eight percent of patients (14/50) acquired at least one AMRO during the hospital stay. MRS detection frequency at admission was 12% (6/50), while acquisition was 6% (3/50). 3GCR-GNB detection frequency was 14% at admission (7/50) and acquisition 22% (11/50), while CR-GNB detection frequency was 2% at admission (1/50) and acquisition 2% (1/50). Environmental surveillance (98 samples) showed a total detection frequency of 22.4% for MRS (22/98), 2% for 3GCR-GNB and CR-GNB (2/98). Clinical staff shoe soles showed high detection frequency for MRS (50%). 3GCR *Escherichia coli* was the most isolated species in patients (n = 17). The results show how active surveillance can be used as a tool to assess the impact of AMROs in veterinary hospitals to subsequently build up tailored control plans based on specific issues.

1. Introduction

Antimicrobial resistance (AMR) is considered the most important emerging threat in both human and veterinary medicine. AMR can be seen as a natural consequence of the antibiotic era due to the selective pressure exerted by antibiotics, but the increasing use (including misuse

or overuse) in the last decades is accelerating the phenomenon at an exponential level [1]. Specifically, hospitals are considered one of the major sources for AMR onset, due to the massive use of antimicrobials and the presence of high-risk patients [2]. Such as human settings, also veterinary hospitals are experiencing the same issue [3] and need further attention to guarantee the healthcare for the animals,

Abbreviations: AMR, antimicrobial resistance; AMRO, antimicrobial-resistant organisms; AST, antimicrobial susceptibility testing; CDT, combination disc test; CR-GNB, carbapenem-resistant gram-negative bacteria; ESBL-E, extended spectrum beta-lactamases producing Enterobacterales; ESCMID, European Society of Clinical Microbiology and Infectious Diseases; EUCAST, European Committee of Antimicrobial Susceptibility Testing; HCAI, healthcare-associated infections; MALDI-TOF, matrix-assisted laser desorption/ionization time of flight; MRS, methicillin-resistant Staphylococci; MRSA, methicillin-resistant *Staphylococcus aureus*; MRSP, methicillin-resistant *Staphylococcus pseudintermedius*; 3GCR-GNB, 3rd generation cephalosporins-resistant gram-negative bacteria; VTH, veterinary teaching hospital.

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considering that the some antimicrobials used are shared, and so the emerging antimicrobial-resistant organisms (AMROs). While hospitalized, patients can be colonized by AMROs in their commensal flora, with subsequent spread to the local community when discharged, or can contract healthcare-associated infections (HCAs). HCAs are often caused by AMROs, and represent a serious threat for their high morbidity and mortality rates, the association with prolonged hospital stays and the significant financial burden [4]. From a global One Health perspective, some AMROs are considered particularly relevant. Methicillin-resistant *Staphylococci* (MRS) are well-recognized as important infectious agents, often showing multi-resistance patterns [5]. While in human medicine the major pathogen MRS is MR *Staphylococcus aureus* (MRSA), in small animal practice MR *Staphylococcus pseudintermedius* (MRSP) is the most relevant species, often associated with skin, surgical site or urinary tract infections [6,7]. The emergence of gram-negative bacteria resistant to third-generation cephalosporins (3GCR-GNB), including Enterobacterales able to produce extended spectrum beta-lactamases (ESBL-E), is another health concern [8]. ESBL production is associated with resistance to penicillins and cephalosporins, and in some cases to beta-lactamase inhibitors [9,10]. Furthermore, despite carbapenems are not approved for veterinary use [11], the onset of carbapenem-resistant gram-negative bacteria (CR-GNB) has also been described in small animal practice [12–14]. In a One Health context, small animal veterinary hospitals should be monitored with attention. Indeed, still little is known about their role in the onset and dissemination of AMROs [15], but they represent an epidemiological point of risk, also in relation with the risk of zoonotic transmission, as previously described by many authors [16–18]. Indeed, dogs and cats are increasingly popular in the family households, especially in high income countries after the COVID-19 pandemic [19,20], and are often at close contact with people, enhancing the chances of AMR transmission.

For these reasons, the implementation of surveillance systems can be a useful and cost-effective tool also for veterinary hospitals [21]. These systems mainly collect, organize, and analyze information subsequently used for the building of tailored infection control policies, including antimicrobial stewardship programs. In this way, surveillance improves not only patient safety, but also reduced the costs related with HCAs outbreaks. Although efforts are in progress, small animal practice still lacks standardized and coordinated procedures, so self-reporting systems could be used by veterinary hospitals as a first step in the process. Surveillance can be executed by collecting and analyzing data already present (passive surveillance) or by actively screening patients (to find asymptomatic carriers of AMROs), the personnel or the environment. In order to minimize the costs, it can be targeted on specific patients (e.g., oncological patients), bacterial species (e.g. MRS) or syndromic events (e.g. fever of unknown origin). The aim of this study is to describe a pilot program of active surveillance developed in a Spanish Veterinary Teaching Hospital (VTH) to estimate the detection frequency of specific AMROs (MRS, 3GCR-GNB, CR-GNB) in the commensal flora of hospitalized patients and in the environment (including hospital personnel), to define where to focus on in long-term prevention and control policies.

2. Materials and methods

2.1. Study design

From October 2022 to February 2023, a longitudinal prospective study was conducted into a Spanish Veterinary Teaching Hospital, as a part of a pilot experimental surveillance program for the assessment of AMR in VTHs. The study included: i) pulsed active surveillance on patients for the evaluation of the commensal flora; ii) active surveillance on environment and staff; and iii) feedback reports.

2.1.1. Pulsed active surveillance on patients

Active surveillance on patients was performed in two periodic sessions (pulsed surveillance), within a month from each other. According

with clinical staff's indications, every patient expected to be hospitalized for more than 48 h was sampled twice, at admission and before discharge, until reaching twenty-five patients for each session. For every patient, demographic data (species, sex) and the length of hospitalization were recorded.

2.1.2. Active surveillance on environment and staff

Environmental surveillance was performed monthly for four times by sampling surfaces (chairs, exam tables, keyboards, stretchers and stretcher wheels, tables and lead gowns of X-ray room, table in the laboratory of bacteriology) and devices (dogs scale, thermometers, urgency phone, hair clipper, portable ultrasound machine) from different areas (waiting room, general wards, consultation room, X-ray room, laboratory of bacteriology). With the same modalities, samples from randomly chosen (simple random sampling) hospital staff were taken. For each person, three samples (hands, clothes and shoe soles) were collected; the same person could be sampled more than once in different months.

2.1.3. Feedback reports

Clinical, surgical, and microbiological personnel was involved in result communication, feedback reports and discussion of single cases through videocalls and chat groups.

2.2. Sampling

Samples were collected with gloved hands and dedicated disposable gowns. Oral and perirectal swabs were collected from the same patients at the admission (within 12 h) and on the day of discharge using sterile swabs with Amies transport medium. Oral sampling was performed by gently inserting the swab for 10–15 seconds into the oral cavity, lateral to the tongue. Perirectal samplings were performed by gently putting the swab in the perirectal area and rotating it for 10–15 s. For environmental surveillance, samples from personnel' hands were collected with sterile swabs with AMIES transport media by scrubbing the swab on the palm and the back of both hands for 5–10 s each. All the other samples (equipment, surfaces and shoe soles) were collected using sterile sponges pre-soaked with 5 ml of sterile saline solution and scrubbed over the target in an area of approximately 80–100 cm². After the sampling, sponges were added with 5 ml of additional sterile solution and squeezed, then the liquid transferred in a sterile tube. Samples were stored at 4 °C for a maximum of 24 h before being processed.

2.3. Isolation and identification

Every sample was then processed by streaking into selective chromogenic media. Perirectal swabs were cultured on CHROMAGAR TM KPC (Chromagar™, Paris, France) for CR-GNB, and on CHROMAGAR TM ESBL (Chromagar™, Paris, France) for 3GCR-GNB. Oral swabs were cultured on Oxacillin Resistance Screening Agar Base (ORSAB, Oxoid, Wesel, Germany) for MRS. Environmental samples were cultured on all the three selective media. Bacterial culture was performed by rubbing the swab over approximately one third of the surface of each plate, and subsequently streaking using a sterile loop. For the samples collected using sterile sponges, the bacterial culture was performed by directly streaking 10 microliters of the liquid in the plate with a sterile loop. After 24–48 h of incubation at 37 °C in aerobic conditions, colonies chromogenically different isolated from positive cultures were sub-cultured on blood agar with 5% sheep blood (Oxoid) and subsequently identified with the matrix-assisted laser desorption/ionization time of flight (MALDI-TOF) using a Bruker Daltonics UltrafleXtreme equipment, and the Biotyper Real Time Classification software v3.1 (Bruker Daltonics, Germany). Interpretation was done according to Bruker standard criteria. Colonies identified with a score > 2.2 were considered at species level, while colonies with a score between 1.8 and 2.2 were considered only at the genus level.

2.4. Resistance detection by phenotypic methods

To confirm the resistance pattern of every isolate, a phenotypic confirmatory test was performed by disc diffusion method. Mueller-Hinton Agar (Oxoid) was inoculated with the tested isolates at standard concentration (0.5 Mc Farland), and different antimicrobials were used to test the resistance patterns. Non-susceptibility to both cefotaxime and ceftazidime was used to confirm 3GC resistance, according to EUCAST guidelines [22] (inhibition zones: < 21 mm for cefotaxime 5 µg, < 22 mm for ceftazidime 10 µg). Furthermore, ESBL production in confirmed 3CGR Enterobacterales was evaluated with a combination disc test (CDT) with cephalosporins (ceftazidime 30 µg, cefepime 30 µg, and cefotaxime 30 µg) alone and in combination with clavulanic acid 10 µg, following EUCAST guidelines [22]. An additional confirmatory phenotypic test to discriminate the expression of AmpC β-lactamases was performed through a CDT with disks containing the cephalosporins (ceftazidime 30 µg, cefepime 30 µg, and cefotaxime 30 µg) and both cloxacillin 200 µg and clavulanic acid 10 µg, according to EUCAST [22]. Carbapenem resistance in screened CR-GNB isolates was tested with the commercially available kit for synergy test “KPC&MBL&OXA-48 disc kit” (Liofilchem, Roseto degli Abruzzi, Italy), while MRS isolates were evaluated with the phenotypic confirmatory disc diffusion test described by EUCAST guidelines (cefoxitin 30 µ disc diffusion test with inhibition zone < 22 mm) [22]. For negative control, strains *S. aureus* ATCC 29213 and *Escherichia coli* ATCC 25922 were used for MRS and for 3GCR/CR GNB, respectively. For positive control, genotypically characterized strains from the VTH internal collection were used.

2.5. Data analysis

Descriptive statistics was performed using Microsoft Excel. In patients, detection frequency at admission was assessed by dividing the number of patients with a positive sample for the total number of patients included, while potential in-hospital acquisition was determined by dividing the number of patients with at least one new AMRO species detected at discharge, by the number of total patients. The correlation between frequency of detection at admission and discharge for each AMRO typology (MRS, 3GCR-GNB, CR-GNB) was evaluated with the chi-square test. Results were considered significant at p value < 0.05.

3. Results

3.1. Active surveillance on patients

A total of 50 patients was sampled in two sessions (25 patients in each session), and a total of 100 samples (both perianal and oral) was collected. Forty-two out of fifty (84%) patients were dogs and 8/50 (16%) cats. 27/50 were males and 23/50 females, while the average length of hospitalization was 3.8 days. A total of 49 investigated AMROs was isolated from 24/50 (48%) patients. Overall frequency of detection for MRS was 16% (8/50), while it was 38% (19/50) for 3GCR-GNB and 6% (3/50) for CR-GNB. All the 14 MRS isolates (100%) were confirmed at the disc diffusion test analysis, while the phenotypically confirmed 3GCR-GNB and CR-GNB were 96.7% (30/31) and 75% (3/4), respectively. Considering confirmed 3GCR-Enterobacterales (n = 29/30), ESBL or AmpC production was confirmed by the CDT for all the isolates. In total, 24% of patients (n = 12/50, 9 dogs and 3 cats) were carriers at admission of at least one of the three investigated AMROs, and 4% (n = 2/50, two dogs) were carriers of two. Patients that acquired at least one of the three investigated AMROs during the hospital stay were 14/50 (28%, 13 dogs and one cat). One patient (1/50, 2%, one dog) acquired two AMROs (3CGR *Klebsiella pneumoniae* and CR *Pseudomonas aeruginosa*).

Positivity rates are shown in Fig. 1. Considering MRS screening, detection frequency was 12% (6/50 patients, 5 dogs and one cat) at admission. At discharge, the same patients were still positive; additionally, two more dogs that were negative at admission acquired MRS during hospitalization. One dog that was positive at admission acquired a new MRS species. In total, 8/50 patients (16%, 7 dogs, one cat) were found to be MRS carriers at discharge, and 3/50 (6%) acquired MRS during hospitalization.

Considering confirmed 3GCR-GNB, detection frequency was 14% (7/50 patients, 6 dogs and 1 cat) at admission. The same patients were positive also at discharge; furthermore, 11 patients negative at admission were found to be positive at discharge. In total, 18/50 patients (36%, 16 dogs and 2 cats) were positive at discharge, and 11/50 patients (22%, 10 dogs, one cat) acquired confirmed 3GCR-GNB during hospitalization.

Considering CR-GNB detection, one cat (1/50, 2%) was positive

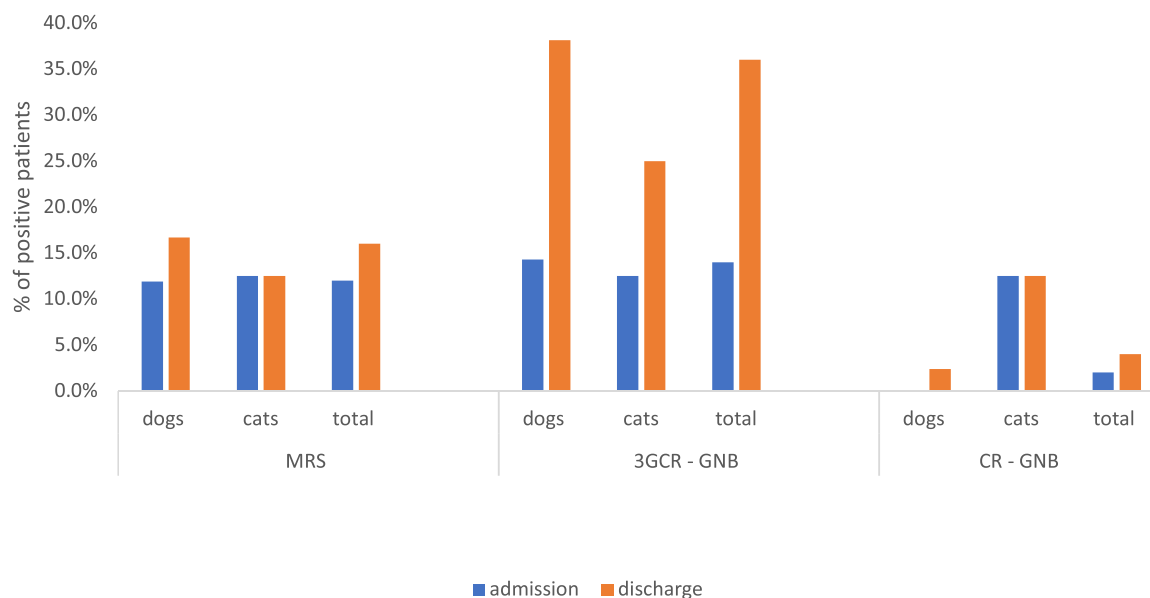


Fig. 1. Detection frequency of MRS, 3GCR-GNB and CR-GNB in the commensal flora of the 50 patients hospitalized for more than 48 h at a VTH and included in the study (from October 2022 to February 2023), sampled with oral and perirectal swabs both at admission (< 12 h) and before discharge. CR-GNB: Carbapenem-resistant gram-negative bacteria. 3GCR-GNB: third-generation cephalosporins resistant gram-negative bacteria. MRS: Methicillin-Resistant Staphylococci. VTH: Veterinary Teaching Hospital.

admission. The same cat was positive also at discharge, and one dog acquired CR-GNB during hospitalization. In total, 2/50 patients (4%) were positive at discharge, and one patient (1/50, 2%) acquired confirmed CR-GNB during hospitalization.

At the statistical analysis, a significant correlation ($p = 0.0115$) was found between 3GCR-GNB detection at admission and at discharge. 3GCR-GNB were more likely to be detected at discharge than at admission.

3.2. Active surveillance on environment and staff

From a total of 98 samples collected, 34 isolates of the investigated AMROs were detected. All the MRS isolates ($n = 22$) were confirmed at the disc diffusion test analysis, while the phenotypically confirmed 3GCR-GNB and CR-GNB were 2/8 and 2/4, respectively. No confirmed 3GCR-Enterobacterales were detected. Total detection frequency was 22.4% for MRS, and 2% for 3GCR-GNB and CR-GNB. Detection frequencies for every sampling site are shown in Table 1. MRS were found to be more present in clinical staff shoe soles (6/12, 50%) and hands (4/12, 33%). 3GCR-GNB and CR-GNB were found in the shoe soles, but also in the hair clipper of the general ward.

3.3. Species identification

Results are shown in Fig. 2. 3GCR-*E. coli* was the most isolated species in patients at admission ($n = 6$) and at discharge ($n = 11$). The three CR-GNB isolated from patients were identified as *S. maltophilia*, *P. aeruginosa* and *K. pneumoniae*. MRSP was the most frequent species found in the environment ($n=8$), followed by MRSA and *Staphylococcus epidermidis* ($n = 4$).

3.4. Feedback reports

A total of three videocalls (one at the beginning of the project, one at the halfway and one at the end) were done to highlight the most critical points, to define how to improve some parts and to discuss single cases. Specifically, we report three anecdotal cases discussed:

- i) During the first session of active surveillance on the environment, of particular concern was the isolation of 3GCR and CR *Acinetobacter pittii* from one of the hair clippers used in the general ward. The result was rapidly communicated, and a specific disinfection method was added for the clipper. This method included a first general cleaning, followed by a disinfection of the blade with 96% alcohol soak for 1 min after every use. Furthermore, the body of the clipper must be disinfected once a day with peroxy-genic acid (Virkon), with a waiting time of 10 min. In the subsequent samplings, no more AMROs were isolated from the clippers.
- ii) During the first session of active surveillance on patients, a cat with an history of previous hospitalizations in a different setting was admitted at the hospital and sampled. The patient had a diagnosed urinary tract infection caused by a multidrug resistant *P. aeruginosa*. The AST from urine performed at the referring clinic showed resistance to all tested antimicrobials except for amikacin, imipenem, and ceftazidime. At the admission, the patient was under treatment with imipenem. After 24 h of incubation, the perirectal sample realized at admission revealed positivity for CR *Stenotrophomonas maltophilia*. The result was communicated to the staff and with the suspect of the development of resistance towards imipenem confirmed by the presence of commensal CR *S. maltophilia* at the perirectal swab, the antimicrobial treatment was changed to ceftazidime, with a rapid improvement of clinical signs.
- iii) A dog with history of chronic skin disease was hospitalized for six days during the first session, and the acquisition of 3GCR-GNB *E. coli* and *K. pneumoniae* during the stay was recorded. During the second session (74 days later), the patient was hospitalized again, and the perirectal swab at the admission revealed the same 3GCR-GNB pattern.

4. Discussion

In a real-life scenario, few veterinary healthcare settings can afford the costs, both in terms of time and money, related with an extensive,

Table 1

Environmental and staff surveillance sampling sites included in the study, and number of positive samples for each site for the detection of MRS, 3GCR-GNB, CR-GNB. Samples were taken monthly from October 2022 to February 2023.

Area	Sampling sites	Total samplings	MRS positives (phenotypically confirmed by CDT)	3GCR-GNB positives (phenotypically confirmed by CDT)	CR-GNB positives (phenotypically confirmed by CDT)
Waiting room	Chairs	4	2	0	0
	Dog scale	2	1	0	0
General Ward	Exam tables	4	1	0	0
	Thermometers	4	0	0	0
	Urgency phone	4	1	0	0
	Keyboards	4	1	0	0
	Hair clipper	4	0	1	1
	Stretcher wheels	4	1	0	0
	Stretcher	4	1	0	0
	Portable ultrasound machine	4	0	0	0
	Clinical staff hands	12	4	0	0
	Clinical staff working clothes	12	1	0	0
Clinical staff shoe soles	12	6	1	1	
Consultation rooms	Exam tables	8	0	0	0
X-Ray room	Lead gowns in X-ray room	4	0	0	0
	Table in X-ray room	4	2	0	0
Laboratory of bacteriology	Laboratory of bacteriology	8	1	0	0
TOTAL		98	22	2	2
% of positive			22.4%	2%	2%

CDT: Combination Disc Test. CR-GNB: Carbapenem-resistant gram-negative bacteria. 3GCR-GNB: third-generation cephalosporins resistant gram-negative bacteria. MRS: Methicillin-Resistant Staphylococci. VTH: Veterinary Teaching Hospital.

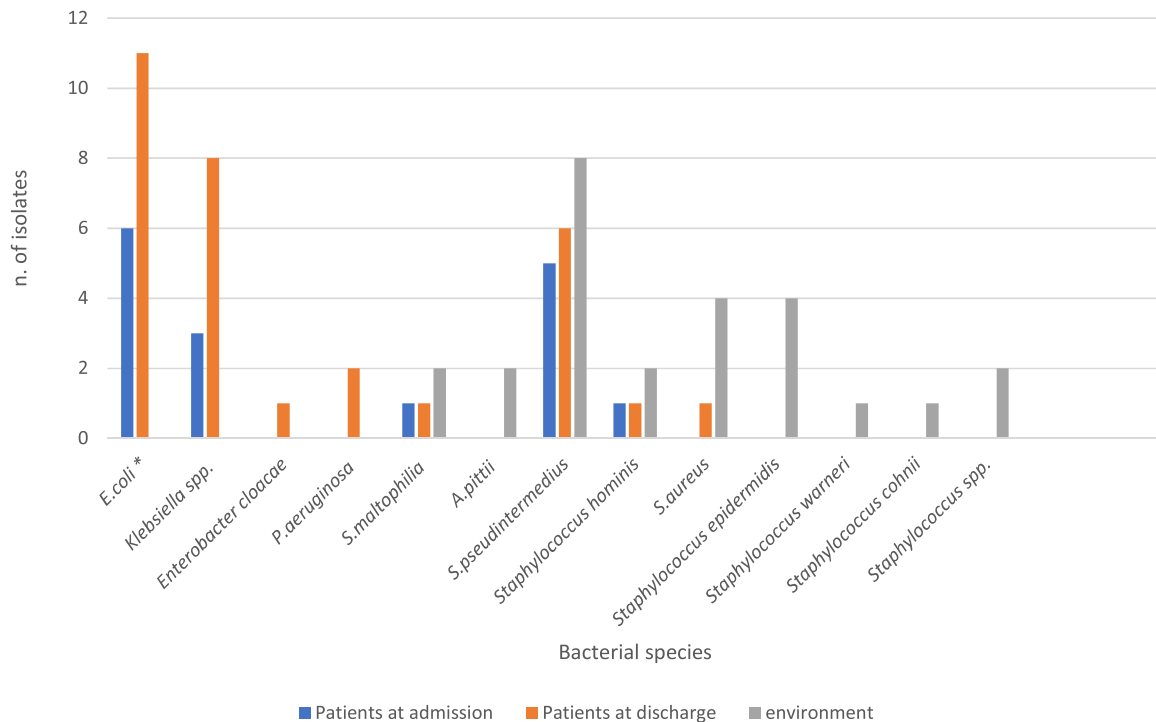


Fig. 2. List of bacterial species isolated and identified with MALDI-TOF, divided considering the isolation site (patients at admission, patients at discharge and environment/staff). *isolates identified as *E. coli* are not distinguishable from *Shigella* spp. with MALDI Biotyper technology.

non-specific, active microbiological surveillance system. For this reason, it becomes necessary to detect where to concentrate the efforts, that should be addressed to the setting-specific needs. In our study, we report the development of a 5-months pilot surveillance program into a Spanish VTH, that aimed to assess the endemic rates of specific AMROs and to identify such priorities.

The results described are comparable with other studies in hospitalized pets from the same region, both at admission and at discharge [23,24]. In Europe, the prevalence of MRS carriage is reported to be generally lower (1.5–9.6%) [25–28], while for 3GCR-GNB it ranges between 1.4% and 45% [26,29–31]. These differences could be due to the different methodologies, patient or targets used, but primarily are attributable to geographical reasons, so comparisons should be done with prudence. Indeed, the general antimicrobial consumption in the local community may primarily influence the results [32]. In Spain, observed prevalence of ESBL-producing Enterobacterales in human patients admitted to hospitals varies between 7.6% and 36.3% [33–35]. At the statistical analysis, a significant correlation was found between 3GCR-GNB positivity at admission and discharge, Considering CR-GNB, the low detection frequency found should not surprise considering that at the time of the samplings carbapenems use was restricted for pets in Spain, and it is in accordance with other studies from the same region [23,36]. Nevertheless, CR-GNB prevalence seems to be increasing in small animal practice and it has been growingly described [14]. Considering that resistance to carbapenems is one of the most concerning public health problems, surveillance of CR-GNB should be enhanced also in veterinary settings to avoid that colonized animals could silently contribute in the dissemination to the community. The confirmatory test for some of the most common carbapenems resistance mechanisms confirmed all except for one isolate (*Klebsiella* spp). A false positive could be possible, but also the production of different resistance mechanisms should be considered [37]. Hence, the absence of a deeper phenotypical analysis is one of the limits of this study.

Comparing dogs and cats, the latter seem to be less susceptible to AMROs acquisition. This finding could have different explanations. First, the lower number of cats involved (8 vs 42 dogs) could have biased

the results. Second, cats tend to have less contact with hospital environment (they are normally brought into the hospital in cat carriers, and do not go out for urinate or defecate): this could lead to a lower exposure to hospital AMROs, and subsequently a lower chance to acquire them. Third, according to studies by Hur et al. [38] and Joosten et al. [39], cats seem to be less frequently treated with antibiotics compared with dogs, and consequently they are less susceptible to AMR. In our case, we did not measure antibiotic treatment in the patients considered, and this consideration cannot be confirmed. Additionally, a statistically significant correlation ($p = 0.0115$) was found between 3GCR-GNB frequency of detection at admission and discharge. The finding suggests that 3GCR-GNB tended to be frequently acquired during hospitalization than MRS or CR-GNB. In companion animals, 3GCR-GNB prevalence seems to be generally higher than MRS and CR-GNB [25–31], with a subsequent higher exposure for susceptible patients to acquire them. Another important factor is the rapid horizontal transmission mechanism that many GNB tend to possess, that leads to a faster and higher acquisition of resistance genes (e.g., ESBL-related genes) compared with MRS, as reported by other studies from the same region [23,24]. On the other hand, CR-GNB acquisition is less likely than 3GCR-GNB due to the absence of selective pressure exerted by carbapenems use.

Screening patients at admission not only provides data about AMROs spread in pets from the local community, but it can be used to reduce the chance of in-hospital transmission in case of a positive carrier. Indeed, through the rapid communication of the results to the clinical personnel, positive patients could be properly managed with extra-preventive measures, such as contact isolation precautions. In human medicine, the 2014 European Society of Clinical Microbiology and Infectious

Diseases (ESCMID) guidelines strongly recommend the implementation of such precautions [40]. Nevertheless, different studies are arguing about the effectiveness of such precautions for ESBL-producing Enterobacterales, due to the absence of evidence-based results able to confirm their ability to reduce in-hospital AMROs transmission and HCAIS in endemic settings [41,42]. The major issues highlighted are the difficulty to afford contact isolation for too many patients and the turnaround times, often too long to allow a proper risk reduction [43,

44]. Furthermore, a universal screening at admission could be difficult to afford in terms of workload and resources [40]. Although specific studies in small animal practice are absent, this topic could be applied also in small animal veterinary hospitals, where often economic resources are scarcer. On the other hand, sampling patients at discharge allows to evaluate the in-hospital AMROs acquisition during time. This can help not only to assess the individual risk of colonization, but also to detect potential epidemic transmissions in carriers before the onset of HCAs. In our case, the results suggest as these screening methods could be focused only on CR-GNB. Indeed, CR-GNB did not seem to be endemically present in the VTH, so the timely detection of positive carriers might be relevant to avoid their spread within the hospital, especially in relation with their importance for public health. The low detection frequency at admission would potentially allow a contact isolation for every positive patient at admission. In addition, although we did not perform any risk factor analysis due to the small population size considered, the screening of carriers at admission should be targeted only on patients with the major risk factors reported in literature, such as previous antimicrobial use or hospitalization, raw meat consumption, long-term use of invasive devices or advanced age [45,46]. The conventional methods to detect AMROs, such as bacteriological culture on selective media, have several limits in terms of time and specificity [43], so then possible, the implementation of faster and more accurate methodologies could be considered to reduce turnaround times or to focus on specific resistance mechanisms.

Environmental contamination of AMROs has been linked with high incidence of HCAs in human medicine [47]. In small animal practice, VTHs are considered a high-risk places due to the high number of referred patients, among others [48], so surveillance on staff and environment is even more relevant. A contaminated environment not only increase the chance to contaminate animals, but also the staff itself, with a potential spread from the hospital to the local community. Walther et al. [49] report that veterinary hospital workers have a major risk of being colonized by AMROs. Through periodic samplings, a hospital-specific risk analysis can be done, allowing to detect the most important hazard points and to develop specific preventive measures, such as the disinfection protocol for the hair clipper described in our case. The results from the samplings in the environment and staff showed a generic discrepancy compared with patients, with a higher detection frequency of MRS (22.4%) compared with 3GCR-GNB (2%), among other facts, this could be due to the low capacity of gram-negative bacteria (especially Enterobacterales such as *E. coli* and *K. pneumoniae*) to survive in the environment [41]. Furthermore, as the highest detection frequencies were related with hospital personnel (shoe soles, hands), their potential role as vectors of resistance should be further investigated. Hand hygiene has been well recognized as an effective factor to reduce HCAs incidence [50]. Indeed, contaminated hands can directly transfer AMROs to patients, but also contaminate fomites and devices, such as the hair clipper. With a proper hand hygiene executed before and after patient contact and contact with high-risk surfaces, the risk of transmission would be reduced [51]. Staff compliance is essential, and can be achieved by direct observation and by measuring the use of alcohol-based hand-rub solution [51,52], or by periodic samplings (e.g. twice a year) of devices and points at higher risk [21]. The importance of shoe soles in the transmission of AMROs have not been well established [52], but the high detection frequency detected suggest they can be the reflection of the floor contamination. Some studies [53–55] have highlighted high prevalence of AMROs in the floor, although a study in an equine hospital [56] described no significant reduction in the floor microbial load after the use of footwear hygiene practice. In small animal practice, patients (especially dogs) tend to have much more contact with the floor, with a subsequent higher exposure, and there is no legislation about minimum cleaning and disinfection standards. Low infection prevention and control standards have been linked with a high AMROs contamination [57]. Veterinary hospitals and large clinics are considered a higher risks compared with

first opinion practices [49], so they should be incentivized to start specific written protocols focused on the primary role of the personnel and on devices reported to have high prevalence.

Feedback reports are an essential part of the surveillance program. Indeed, the continuous, evidence-based result communication to the clinical staff, including about their compliance, can enhance their awareness about the problem, and help to detect the most common practice deficiencies [58,59]. In our case, the three anecdotal examples explain how these feedback reports can be used in an every-day situation.

This study has several limitations. First, although some phenotypical confirmatory tests were performed, the absence of genotypical confirmatory tests did not allow to assess the real presence of resistance genes and a deeper epidemiological investigation. Another limit is the absence of a statistical risk factor analysis for the carriage and the acquisition of AMROs, that could have been implemented for future studies to assess if there were setting-specific risk factors.

5. Conclusions

In conclusion, this study aimed to show how a pilot surveillance study can be used in the first place to understand how to build up a tailored plan based on specific issues. It did not demonstrate the effectiveness of surveillance systems in the reduction of AMROs impact, but describes how large veterinary settings can approach the topic, with an initial plan built to assess the risks and indicate where to concentrate the efforts. Active surveillance should be combined with passive surveillance and tailored antimicrobial stewardship policies, considering that also selective pressure from antimicrobial use plays a major role in the AMROs dissemination. Local data from settings like veterinary hospitals are very important to improve the quality of the global overview about AMR.

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Ethical statement

Ethical approval was obtained for this study from the Ethical Committee of the Veterinary Faculty (ref. n.15/2022, 19/10/2022). Patient's owners gave written consent for the sampling.

CRediT authorship contribution statement

Marta Perez Sancho: Writing – review & editing, Visualization, Validation, Project administration, Methodology, Formal analysis, Data curation. **Miriam Portero Fuentes:** Visualization, Resources, Investigation. **José Luis Blanco:** Writing – review & editing, Visualization, Validation, Supervision, Resources, Project administration, Formal analysis, Conceptualization. **Marta Eulalia Garcia:** Writing – review & editing, Supervision, Resources, Project administration, Conceptualization. **Laura Leal Vélez De Mendizábal:** Writing – review & editing, Methodology, Investigation. **Sergio Quevedo Caraballo:** Investigation. **Silvia Piva:** Writing – review & editing, Validation, Supervision, Resources, Funding acquisition. **Raffaele Scarpellini:** Writing – review & editing, Writing – original draft, Project administration, Methodology, Investigation, Data curation, Conceptualization. **Erika Esposito:** Data curation, Conceptualization. **Elisabetta Mondo:** Visualization. **Silvia Penelo:** Resources, Investigation.

Declaration of Competing Interest

All the authors disclose any financial and personal relationships with other people or organizations that could inappropriately influence (bias)

the work. They all declare that there's no financial/personal interest or belief that could affect their objectivity.

Data availability

Data used for this study are available under request to the corresponding author.

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References

- [1] G. Mancuso, A. Midiri, E. Gerace, C. Biondo, Bacterial antibiotic resistance: the most critical pathogens, *Pathogens* 10 (2021) 1310, <https://doi.org/10.3390/pathogens10101310>.
- [2] K.R. Chng, C. Li, D. Bertrand, A.H.Q. Ng, J.S. Kwah, H.M. Low, C. Tong, M. Natrajan, M.H. Zhang, L. Xu, K.K.K. Ko, E.X.P. Ho, T.V. Av-Shalom, J.W.P. Teo, C.C. Khor, MetaS.U.B. Consortium, D. Danko, D. Bezdán, E. Afshinnekoo, S. Ahsanuddin, C. Bhattacharya, D.J. Butler, K.R. Chng, F. De Filippis, J. Hecht, A. Kahles, M. Karasikov, N.C. Kyripides, M.H.Y. Leung, D. Meleshko, H. Mustafa, B. Mutai, R.Y. Neches, A. Ng, M. Nieto-Caballero, O. Nikolayeva, T. Nikolayeva, E. Png, J.L. Sanchez, H. Shaaban, M.A. Sierra, X. Tong, B. Young, J. Alicea, M. Bhattacharyya, R. Blekhan, E. Castro-Nallar, A.M. Cañas, A. D. Chatziefthimiou, R.W. Crawford, Y. Deng, C. Desnues, E. Dias-Neto, D. Donnellan, M. Dybwad, E. Elhaik, D. Ercolini, A. Frolova, A.B. Graf, D.C. Green, I. Hajirasouliha, M. Hernandez, G. Iraola, S. Jang, A. Jones, F.J. Kelly, K. Knights, P.P. Łabaj, P.K.H. Lee, L. Shawn, P. Ljungdahl, A. Lyons, G. Mason-Buck, K. McGrath, E.F. Mongodin, M.O. Moraes, N. Nagarajan, H. Noushmehr, M. Oliveira, S. Ossowski, O.O. Osualale, O. Özcan, D. Paez-Espino, N. Rascovan, H. Richard, G. Rättsch, L.M. Schriml, T. Semmler, O.U. Sezerman, L. Shi, L.H. Song, H. Suzuki, D.S. Court, D. Thomas, S.W. Tighe, K.I. Udekwo, J.A. Ugalde, B. Valentine, D.I. Vassilev, E. Vayndorf, T.P. Velavan, M.M. Zambrano, J. Zhu, S. Zhu, C.E. Mason, S.L. Chen, C.E. Mason, O.T. Ng, K. Marimuthu, B. Ang, N. Nagarajan, Cartography of opportunistic pathogens and antibiotic resistance genes in a tertiary hospital environment, *Nat. Med.* 26 (2020) 941–951, <https://doi.org/10.1038/s41591-020-0894-4>.
- [3] B. Walther, K. Tedin, A. Lübke-Becker, Multidrug-resistant opportunistic pathogens challenging veterinary infection control, *Vet. Microbiol.* 200 (2017) 71–78, <https://doi.org/10.1016/j.vetmic.2016.05.017>.
- [4] L. Guardabassi, Veterinary hospital-acquired infections: the challenge of MRSA and other multidrug-resistant bacterial infections in veterinary medicine, *Vet. J.* 193 (2012) 307–308, <https://doi.org/10.1016/j.vetj.2012.04.005>.
- [5] K.A. Worthing, J. Brown, L. Gerber, D.J. Trott, S. Abraham, J.M. Norris, Methicillin-resistant staphylococci amongst veterinary personnel, personnel-owned pets, patients and the hospital environment of two small animal veterinary hospitals, *Vet. Microbiol.* 223 (2018) 79–85, <https://doi.org/10.1016/j.vetmic.2018.07.021>.
- [6] V. Perreten, K. Kadlec, S. Schwarz, U. Gronlund Andersson, M. Finn, C. Greko, A. Moodley, S.A. Kania, L.A. Frank, D.A. Bemis, A. Franco, M. Iurescia, A. Battisti, B. Duim, J.A. Wagenaar, E. Van Duijkeren, J.S. Weese, J.R. Fitzgerald, A. Rossano, L. Guardabassi, Clonal spread of methicillin-resistant *Staphylococcus pseudintermedius* in Europe and North America: an international multicentre study, *J. Antimicrob. Chemother.* 65 (2010) 1145–1154, <https://doi.org/10.1093/jac/dkq078>.
- [7] B. Duim, K.M.H.W. Verstappen, R.S. Kalupahana, L. Ranathunga, A.C. Fluit, J. A. Wagenaar, Methicillin-resistant *Staphylococcus pseudintermedius* among dogs in the description of novel SCCmec variants, *Vet. Microbiol.* 213 (2018) 136–141, <https://doi.org/10.1016/j.vetmic.2017.11.022>.
- [8] C.-H. Tseng, C.-W. Liu, P.-Y. Liu, Extended-spectrum β -lactamases (ESBL) producing bacteria in animals, *Antibiotics* 12 (2023) 661, <https://doi.org/10.3390/antibiotics12040661>.
- [9] K. Bush, G.A. Jacoby, Updated functional classification of β -lactamases, *Antimicrob. Agents Chemother.* 54 (2010) 969–976, <https://doi.org/10.1128/AAC.01009-09>.
- [10] D. Rawat, D. Nair, Extended-spectrum β -lactamases in gram negative bacteria, *J. Glob. Infect. Dis.* 2 (2010) 263, <https://doi.org/10.4103/0974-777X.68531>.
- [11] A. Smith, A.S. Wayne, C.L. Fellman, M.H. Rosenbaum, Usage patterns of carbapenem antimicrobials in dogs and cats at a veterinary tertiary care hospital, *Vet. Intern. Med.* 33 (2019) 1677–1685, <https://doi.org/10.1111/jvim.15522>.
- [12] A. Nigg, M. Brillhante, V. Dazio, M. Clément, A. Collaud, S. Gobeli Brawand, B. Willi, A. Endimiani, S. Schuller, V. Perreten, Shedding of OXA-181 carbapenemase-producing *Escherichia coli* from companion animals after hospitalisation in Switzerland: an outbreak in 2018, *Eurosurveillance* 24 (2019), <https://doi.org/10.2807/1560-7917.ES.2019.24.39.1900071>.
- [13] S.D. Cole, L. Peak, G.H. Tyson, R. Reimschuessel, O. Ceric, S.C. Rankin, New Delhi metallo- β -lactamase-5-producing *Escherichia coli* in companion animals, *United States, Emerg. Infect. Dis.* 26 (2020) 381–383, <https://doi.org/10.3201/eid2602.191221>.
- [14] J.M.D. Silva, J. Menezes, C. Marques, C.F. Pomba, Companion animals—an overlooked and misdiagnosed reservoir of carbapenem resistance, *Antibiotics* 11 (2022) 533, <https://doi.org/10.3390/antibiotics11040533>.
- [15] D.C. Sebola, J.W. Oguttu, M.M. Kock, D.N. Qekwana, Hospital-acquired and zoonotic bacteria from a veterinary hospital and their associated antimicrobial-susceptibility profiles: a systematic review, *Front. Vet. Sci.* 9 (2023) 1087052, <https://doi.org/10.3389/fvets.2022.1087052>.
- [16] T. Grönthal, M. Österblad, M. Eklund, J. Jalava, S. Nykäsenoja, K. Pekkanen, M. Rantala, Sharing more than friendship – transmission of NDM-5 ST167 and CTX-M-9 ST69 *Escherichia coli* between dogs and humans in a family, *Finland, 2015, Eurosurveillance* 23 (2018), <https://doi.org/10.2807/1560-7917.ES.2018.23.27.1700497>.
- [17] J.E. Rubin, J.D.D. Pitout, Extended-spectrum β -lactamase, carbapenemase and AmpC producing Enterobacteriaceae in companion animals, *Vet. Microbiol.* 170 (2014) 10–18, <https://doi.org/10.1016/j.vetmic.2014.01.017>.
- [18] R. Somayaji, M.A.R. Priyantha, J.E. Rubin, D. Church, Human infections due to *Staphylococcus pseudintermedius*, an emerging zoonosis of canine origin: report of 24 cases, *Diagn. Microbiol. Infect. Dis.* 85 (2016) 471–476, <https://doi.org/10.1016/j.diagmicrobio.2016.05.008>.
- [19] J. Ho, S. Hussain, O. Sparagano, Did the COVID-19 pandemic spark a public interest in pet adoption? *Front. Vet. Sci.* 8 (2021) 647308, <https://doi.org/10.3389/fvets.2021.647308>.
- [20] C.L. Hoffman, M. Thibault, J. Hong, Characterizing Pet acquisition and retention during the COVID-19 pandemic, *Front. Vet. Sci.* 8 (2021) 781403, <https://doi.org/10.3389/fvets.2021.781403>.
- [21] B.A. Burgess, P.S. Morley, Veterinary hospital surveillance systems, *Vet. Clin. N. Am.: Small Anim. Pract.* 45 (2015) 235–242, <https://doi.org/10.1016/j.cvsm.2014.11.002>.
- [22] European Committee on Antimicrobial Susceptibility Testing (EUCAST), EUCAST guidelines for detection of resistance mechanisms and specific resistances of clinical and/or epidemiological importance – Version 2.0., EUCAST, Växjö, 2017. (https://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Resistance_mechanisms/EUCAST_detection_of_resistance_mechanisms_170711.pdf).
- [23] G. Ortiz-Díez, R.L. Mengíbar, M.-C. Turrientes, M.-R.B. Artigao, R.L. Gallifa, A. M. Tello, C.F. Pérez, T.A. Santiago, Prevalence, incidence and risk factors for acquisition and colonization of extended-spectrum beta-lactamase- and carbapenemase-producing Enterobacteriaceae from dogs attended at a veterinary hospital in Spain, *Comp. Immunol. Microbiol. Infect. Dis.* 92 (2023) 101922, <https://doi.org/10.1016/j.cimid.2022.101922>.
- [24] G. Ortiz-Díez, R. López, A.M. Sánchez-Díaz, M.-C. Turrientes, M.-R. Baquero, R. Luque, A. Maroto, C. Fernández, T. Ayllón, Epidemiology of the colonization and acquisition of methicillin-resistant staphylococci and vancomycin-resistant enterococci in dogs hospitalized in a clinic veterinary hospital in Spain, *Comp. Immunol. Microbiol. Infect. Dis.* 72 (2020) 101501, <https://doi.org/10.1016/j.cimid.2020.101501>.
- [25] V. Dazio, A. Nigg, J.S. Schmidt, M. Brillhante, N. Mauri, S.P. Kuster, S.G. Brawand, G. Schüpbach-Regula, B. Willi, A. Endimiani, V. Perreten, S. Schuller, Acquisition and carriage of multidrug-resistant organisms in dogs and cats presented to small animal practices and clinics in Switzerland, *Vet. Intern. Med.* 35 (2021) 970–979, <https://doi.org/10.1111/jvim.16038>.
- [26] U. Kaspar, A. Von Lützu, A. Schlattmann, U. Roesler, R. Köck, K. Becker, Zoonotic multidrug-resistant microorganisms among small companion animals in Germany, *PLoS One* 13 (2018) e0208364, <https://doi.org/10.1371/journal.pone.0208364>.
- [27] E. Hamilton, J.M. Kruger, W. Schall, M. Beal, S.D. Manning, J.B. Kaneene, Acquisition and persistence of antimicrobial-resistant bacteria isolated from dogs and cats admitted to a veterinary teaching hospital, *J. Am. Vet. Med. Assoc.* 243 (2013) 990–1000, <https://doi.org/10.2460/javma.243.7.990>.
- [28] A. Bergström, C. Gustafsson, M. Leander, M. Fredriksson, U. Grönlund, G. Trowald-Wigh, Occurrence of methicillin-resistant *Staphylococci* in surgically treated dogs and the environment in a Swedish animal hospital, *J. Small Anim. Pract.* 53 (2012) 404–410, <https://doi.org/10.1111/j.1748-5827.2012.01238.x>.
- [29] J. Hordijk, A. Schoormans, M. Kwakernaak, B. Duim, E. Broens, C. Dierikx, D. Mevius, J.A. Wagenaar, High prevalence of fecal carriage of extended spectrum β -lactamase/AmpC-producing Enterobacteriaceae in cats and dogs, *Front. Microbiol.* 4 (2013), <https://doi.org/10.3389/fmicb.2013.00242>.
- [30] A.L. Wedley, S. Dawson, T.W. Maddox, K.P. Coyne, G.L. Pinchbeck, P. Clegg, T. Nuttall, M. Kirchner, N.J. Williams, Carriage of antimicrobial resistant *Escherichia coli* in dogs: prevalence, associated risk factors and molecular characteristics, *Vet. Microbiol.* 199 (2017) 23–30, <https://doi.org/10.1016/j.vetmic.2016.11.017>.
- [31] G. van den Bunt, A.C. Fluit, M.P. Spaninks, A.J. Timmerman, Y. Geurts, A. Kant, J. Scharinga, D. Mevius, J.A. Wagenaar, M.J.M. Bonten, W. van Pelt, J. Hordijk, Faecal carriage, risk factors, acquisition and persistence of ESBL-producing Enterobacteriaceae in dogs and cats and co-carriage with humans belonging to the same household, *J. Antimicrob. Chemother.* 75 (2020) 342–350, <https://doi.org/10.1093/jac/dkz462>.
- [32] C. Manyi-Loh, S. Mamphweli, E. Meyer, A. Okoh, Antibiotic use in agriculture and its consequential resistance in environmental sources: potential public health implications, *Molecules* 23 (2018) 795, <https://doi.org/10.3390/molecules23040795>.
- [33] C. Díaz-Agero Pérez, N. López-Fresneña, A.L. Rincon Carlavilla, M. Hernandez Garcia, P. Ruiz-Garabaja, J.M. Aranz-Andrés, F. Maechler, P. Gastmeier, M.J. M. Bonten, R. Canton, Local prevalence of extended-spectrum beta-lactamase (ESBL) producing Enterobacteriaceae intestinal carriers at admission and co-

- expression of ESBL and OXA-48 carbapenemase in *Klebsiella pneumoniae*: a prevalence survey in a Spanish University Hospital, *BMJ Open* 9 (2019) e024879, <https://doi.org/10.1136/bmjopen-2018-024879>.
- [34] C. Colmenero, M. Hernández-García, J.R. Muñoz-Rodríguez, N. Huertas, F. J. Navarro, A.B. Mateo, E.M. Pellejero, S. Illescas, M.D. Vidal, R. Del Campo, Prevalence and risks factors associated with ESBL-producing faecal carriage in a single long-term-care facility in Spain: emergence of CTX-M-24- and CTX-M-27-producing *Escherichia coli* ST131-H30R, *J. Antimicrob. Chemother.* 75 (2020) 2480–2484, <https://doi.org/10.1093/jac/dkaa219>.
- [35] C. Del Rosario-Quintana, T. Tosco-Núñez, L. Lorenzo, A.M. Martín-Sánchez, J. Molina-Cabrillana, Prevalencia y factores asociados a la colonización de microorganismos multirresistentes en centros de larga estancia de Gran Canaria, *Rev. Española Geriatr. y Gerontol.* 50 (2015) 232–236, <https://doi.org/10.1016/j.regg.2014.11.006>.
- [36] A. González-Torralba, J. Oteo, A. Asenjo, V. Bautista, E. Fuentes, J.-I. Alós, Survey of carbapenemase-producing enterobacteriaceae in companion dogs in Madrid, Spain, *Antimicrob. Agents Chemother.* 60 (2016) 2499–2501, <https://doi.org/10.1128/AAC.02383-15>.
- [37] T. Karampatakis, K. Tsergouli, P. Behzadi, Carbapenem-Resistant *Klebsiella pneumoniae*: virulence factors, molecular epidemiology and latest updates in treatment options, *Antibiotics* 12 (2023) 234, <https://doi.org/10.3390/antibiotics12020234>.
- [38] B.A. Hur, L.Y. Hardefeldt, K.M. Verspoor, T. Baldwin, J.R. Gilkerson, Describing the antimicrobial usage patterns of companion animal veterinary practices; free text analysis of more than 4.4 million consultation records, *PLoS One* 15 (2020) e0230049, <https://doi.org/10.1371/journal.pone.0230049>.
- [39] P. Joosten, D. Ceccarelli, E. Odent, S. Sarrazin, H. Graveland, L. Van Gompel, A. Battisti, A. Caprioli, A. Franco, J.A. Wagenaar, D. Mevius, J. Dewulf, Antimicrobial usage and resistance in companion animals: a cross-sectional study in three European countries, *Antibiotics* 9 (2020) 87, <https://doi.org/10.3390/antibiotics9020087>.
- [40] E. Tacconelli, M.A. Cataldo, S.J. Dancer, G. De Angelis, M. Falcone, U. Frank, G. Kahlmeter, A. Pan, N. Petrosillo, J. Rodríguez-Baño, N. Singh, M. Venditti, D. S. Yokoe, B. Cookson, ESCMID guidelines for the management of the infection control measures to reduce transmission of multidrug-resistant Gram-negative bacteria in hospitalized patients, *Clin. Microbiol. Infect.* 20 (2014) 1–55, <https://doi.org/10.1111/1469-0691.12427>.
- [41] S. Tschudin-Sutter, J.-C. Lucet, N.T. Mutters, E. Tacconelli, J.R. Zahar, S. Harbarth, Contact precautions for preventing nosocomial transmission of extended-spectrum β lactamase-producing *Escherichia coli*: a point/counterpoint review, *Clin. Infect. Dis.* 65 (2017) 342–347, <https://doi.org/10.1093/cid/cix258>.
- [42] F. Maechler, F. Schwab, S. Hansen, C. Fankhauser, S. Harbarth, B.D. Huttner, C. Diaz-Agero, N. Lopez, R. Canton, P. Ruiz-Garbajosa, H. Blok, M.J. Bonten, F. Kloosterman, J. Schotsman, B.S. Cooper, M. Behnke, J. Golembus, A. Kola, P. Gastmeier, Contact isolation versus standard precautions to decrease acquisition of extended-spectrum β -lactamase-producing Enterobacterales in non-critical care wards: a cluster-randomised crossover trial, *Lancet Infect. Dis.* 20 (2020) 575–584, [https://doi.org/10.1016/S1473-3099\(19\)30626-7](https://doi.org/10.1016/S1473-3099(19)30626-7).
- [43] D.J. Diekema, M.A. Pfaller, Rapid detection of antibiotic-resistant organism carriage for infection prevention, *Clin. Infect. Dis.* 56 (2013) 1614–1620, <https://doi.org/10.1093/cid/cit038>.
- [44] F. Maechler, F. Schwab, S. Hansen, M. Behnke, M.J. Bonten, R. Canton, C. Diaz Agero, C. Fankhauser, S. Harbarth, B.D. Huttner, A. Kola, P. Gastmeier, Quantification of time delay between screening and subsequent initiation of contact isolation for carriers of extended-spectrum beta-lactamase (ESBL)-producing Enterobacterales: a post hoc subgroup analysis of the R-GNOSIS WP5 Trial, *Infect. Control Hosp. Epidemiol.* (2023) 1–7, <https://doi.org/10.1017/ice.2022.285>.
- [45] C. Pomba, M. Rantala, C. Greko, K.E. Baptiste, B. Catry, E. van Duijkeren, A. Mateus, M.A. Moreno, S. Pyörälä, M. Ružauskas, P. Sanders, C. Teale, E. J. Threlfall, Z. Kunsagi, J. Torren-Edo, H. Jukes, K. Törneke, Public health risk of antimicrobial resistance transfer from companion animals, *J. Antimicrob. Chemother.* (2016) dkw481, <https://doi.org/10.1093/jac/dkw481>.
- [46] M. Nüesch-Inderbinen, A. Treier, K. Zurlfluh, R. Stephan, Raw meat-based diets for companion animals: a potential source of transmission of pathogenic and antimicrobial-resistant Enterobacteriaceae, *R. Soc. Open Sci.* 6 (2019) 191170, <https://doi.org/10.1098/rsos.191170>.
- [47] E. Odoyo, D. Matano, F. Tiria, M. Georges, C. Kyanya, S. Wahome, W. Mutai, L. Musila, Environmental contamination across multiple hospital departments with multidrug-resistant bacteria pose an elevated risk of healthcare-associated infections in Kenyan hospitals, *Antimicrob. Resist. Infect. Control* 12 (2023) 22, <https://doi.org/10.1186/s13756-023-01227-x>.
- [48] K.M. Benedict, P.S. Morley, D.C.V. Metre, Characteristics of biosecurity and infection control programs at veterinary teaching hospitals, *J. Am. Vet. Med. Assoc.* 233 (2008) 767–773, <https://doi.org/10.2460/javma.233.5.767>.
- [49] B. Walther, K. Tedin, A. Lübke-Becker, Multidrug-resistant opportunistic pathogens challenging veterinary infection control, *Vet. Microbiol.* 200 (2017) 71–78, <https://doi.org/10.1016/j.vetmic.2016.05.017>.
- [50] B. Allegranzi, D. Pittet, Role of hand hygiene in healthcare-associated infection prevention, *J. Hosp. Infect.* 73 (2009) 305–315, <https://doi.org/10.1016/j.jhin.2009.04.019>.
- [51] T. Julian, A. Singh, J. Rousseau, J.S. Weese, Methicillin-resistant staphylococcal contamination of cellular phones of personnel in a veterinary teaching hospital, *BMC Res. Notes* 5 (2012) 193, <https://doi.org/10.1186/1756-0500-5-193>.
- [52] J. Wojtacka, B. Wysok, A. Kocuvan, M. Rupnik, High contamination rates of shoes of veterinarians, veterinary support staff and veterinary students with *Clostridioides difficile* spores, *Transboundary Emerg. Dis.* 69 (2022) 685–693, <https://doi.org/10.1111/tbed.14034>.
- [53] E. Aksoy, A. Boag, D. Brodbelt, J. Grierson, Evaluation of surface contamination with staphylococci in a veterinary hospital using a quantitative microbiological method, *J. Small Anim. Pract.* 51 (2010) 574–580, <https://doi.org/10.1111/j.1748-5827.2010.00994.x>.
- [54] P. Fungwithaya, J. Murugaiyan, N. Prapasarakul, Distribution and correlation of methicillin-resistant coagulase-positive staphylococci (MRCoPS) between environmental surfaces, veterinary staff and dogs within a veterinary teaching hospital, Thailand, In Review, 2021. (<https://doi.org/10.21203/rs.3.rs-244889/v1>).
- [55] J. van Balen, C. Kelley, R.C. Nava-Hoet, S. Bateman, A. Hillier, J. Dyce, T. E. Wittum, A.E. Hoet, Presence, distribution, and molecular epidemiology of methicillin-resistant *Staphylococcus aureus* in a small animal teaching hospital: a year-long active surveillance targeting dogs and their environment, *Vector-Borne Zoonotic Dis.* 13 (2013) 299–311, <https://doi.org/10.1089/vbz.2012.1142>.
- [56] K.A. Stockton, P.S. Morley, D.R. Hyatt, B.A. Burgess, G. Patterson, M. Dunowska, D. E. Lee, Evaluation of the effects of footwear hygiene protocols on nonspecific bacterial contamination of floor surfaces in an equine hospital, *Javma* 228 (2006) 1068–1073, <https://doi.org/10.2460/javma.228.7.1068>.
- [57] J.S. Schmidt, S.P. Kuster, A. Nigg, V. Dazio, M. Brilhante, H. Rohrbach, O. J. Bernasconi, T. Büdel, E.I. Campos-Madueno, S. Gobeli Brawand, S. Schuller, A. Endimiani, V. Perreten, B. Willi, Poor infection prevention and control standards are associated with environmental contamination with carbapenemase-producing Enterobacterales and other multidrug-resistant bacteria in Swiss companion animal clinics, *Antimicrob. Resist. Infect. Control* 9 (2020) 93, <https://doi.org/10.1186/s13756-020-00742-5>.
- [58] T. Wu, L. Jie, M.T.M. Cabahug, P. Liew, C. Hairu, S.B. Nasir, H. Kaur, A. Rongyan, F.S. Yun, T.S. Yen, T.T. Yen, T.S. Huei, SG-APSIC1159: control of hospital-acquired carbapenemase-producing carbapenem-resistant Enterobacteriaceae colonization: a descriptive study, *ASHE* 3 (2023) s27–s28, <https://doi.org/10.1017/ash.2023.82>.
- [59] A.W. Baker, I. Ilies, J.C. Benneyan, Y. Lokhnygina, K.R. Foy, S.S. Lewis, B. Wood, E. Baker, L. Crane, K.L. Crawford, A.L. Cromer, P. Padgett, L. Roach, L. Adcock, N. Nehls, J. Salem, D. Bratzler, E.P. Dellinger, L.R. Greene, S.S. Huang, C. R. Mantyh, D.J. Anderson, Early recognition and response to increases in surgical site infections using optimised statistical process control charts—the early 2RIS trial: a multicentre stepped wedge cluster randomised controlled trial, *eClinicalMedicine* 54 (2022) 101698, <https://doi.org/10.1016/j.eclinm.2022.101698>.