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A PEEK BEHIND THE CURTAIN ON ESBL-MDR *KLEBSIELLA PNEUMONIAE* AND *ESCHERICHIA COLI* AMONG PET ANIMALS SUFFERING FROM OTITIS

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ABSTRACT

The current study was conducted to investigate ear canal colonization by ESBL-KP and ESBL-EC in pets clinically diagnosed as infectious otitis cases. Also, predisposition correlation links between the isolation rate and several related statuses, such as age, habitat, and breed of the incorporated cases. Ear swabs were obtained from 118 and 94 pet dogs and cats, respectively, with no known history of hospitalization. All samples were enriched before being cultured on the selective MacConkey agar and the enriched 5% sheep blood agar to isolate the Enterobacteriaceae members of interest. *K. pneumoniae* and *E. coli* were further identified by biochemical and molecular techniques. Antimicrobial resistance profiles of the obtained isolates were determined by the Kirby Bauer method, and the multi-drug-resistant strains were examined by PCR to detect the *bla*CTX-M and *bla*TEM genes. Ear prevalence rates of ESBL-KP among pet dogs and cats were 17.8% and 18%, respectively, whilst those for ESBL-EC were 12.4% and 9.6%, respectively. None of the isolated *K. pneumoniae* and *E. coli* were sensitive to linezolid, tylosin, and lincomycin. However, *K. pneumoniae* and *E. coli* resistant to ceftriaxone, ceftazidime, and cefoperazone were detected in the examined samples at rates of 17.6%, 19.4%, and 21.2%, respectively. Moreover, among the isolated *K. pneumoniae* and *E. coli*, all obtained isolates showed multidrug resistance. The *bla*CTX-M and *bla*TEM genes were detected on all canine and feline MDR isolates. The occurrence of *bla*CTX-M and *bla*TEM genes highlights the role of pet animals as a possible source of transmission of such pathogens to humans.

Keywords: *bla*CTX-M gene; *bla*TEM gene; ESBL-KP and ESBL-EC; MDR; WHO GLASS

INTRODUCTION

A growing public health concern, which must be considered in a One Health perspective, is antimicrobial resistance (AMR), which has been described as “the silent tsunami facing modern medicine” (Aslam *et al*., 2021). Since the food chain is recognized to be a significant pathway for the spread of infectious agents carrying AMR features, surveillance programs for One Health AMR primarily focus on the elements that make up the food chain (Depoorter *et al*., 2012). The food chain is also the focus of the World Health Organization's (WHO) proposed Tricycle Global Antimicrobial Resistance Surveillance System (GLASS), which includes pregnant women in the community

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for the human sector, live chickens in open markets for the animal sector, and the river near the abattoir for the environmental sector (WHO, 2021).

Following the WHO Advisory Group on Integrated Surveillance on AMR's recommendation, CTX-M-type extended-spectrum beta-lactamase (ESBL)-producing Escherichia coli (ESBL-EC) was included in the GLASS as a sentinel AMR organism. The argument was based on (I) different rates of human colonization of ESBL-EC within and between nations (Woerther *et al*., 2013); (II) varying prevalence among farm animals and some evidence that a portion of the human morbidity associated with ESBL-ECs is caused by both the environmental presence (Mughini-Gras *et al*., 2019) of ESBL-ECs and the use of antibiotics in the food chain (ECDC, 2019); (III) interventions to reduce antimicrobial exposure in animals (Korsgaard *et al*., 2020) or humans (Schechner *et al*., 2013) led to a decrease in the rates of occurrence of ESBL-Ecs; and (IV) ESBL production by the infection-caused pathogens restricts treatment options (Xu *et al*., 2017), resulting in the use of last resort medications (Pitout *et al*., 2015).

Certain bacterial species must be selected for the surveillance study for certain animals, as the protocol said, because the sentinel bacterial species with the designated AMR feature is not representative and insufficient to understand all. Additionally, different bacterial species may have different dissemination mechanisms. For example, ESBL-producing *K. pneumoniae* (ESBL-KP) spread was mostly caused by clonal dispersion across patients, whereas ESBL-EC spread was caused by horizontal gene transfer (Dural *et al*., 2019).

Since pet animals live with humans both indoors and outdoors, they are an essential part of the animal sector, even though they are not part of the food chain (Orengaauw *et al*., 2020; Miller, 2018). Direct and indirect interactions between humans and pet animals can provide an opportunity for the spread of AMR. The position of pet animals in the present One Health AMR framework is highlighted by the fact that MDR bacteria from pet animals have been reported frequently worldwide (Cui *et al*., 2018; Menezes *et al*., 2023). Additionally, veterinarians frequently treat companion animals with infectious diseases using antimicrobial medications reserved for humans (Overgaauw *et al*., 2020). This can result in selection pressure that promotes intra- and inter-sectoral dissemination, either by horizontal gene transfer of AMR genes to bacterial pathogens or by clonal dissemination (Holmes *et al*., 2016).

In this context, the rise of multidrug resistance in Gram-negative bacteria (MDR-GNB) has become a particularly serious challenge on both the national and international levels, specifically AMR in a list of common enteric bacteria, which may cause serious and different infectious diseases in a wide range of hosts. ESBL-EC and ESBL-KP at the top of the list are considered the most critical group of pathogens posing a considerable threat to human health (WHO, 2017).

Many studies have focused on the detection of ESBL in swine, poultry and food-producing animals, but the role of animals in the epidemiology of ESBL-EC and ESBL-KP remains poorly understood. Moreover, the occurrence of ESBL-EC and ESBL-KP among companion animals has been reported previously. Nonetheless, there is a scarcity of data on the occurrence of such pathogens in the otitis cases of pet dogs and cats. Therefore, the present study was conducted to investigate and evaluate the occurrence of ESBL-EC and ESBL-KP multidrug-resistant pathogens in the otitis cases of pet animals.

MATERIALS AND METHODS

Ethics statement

All dogs and cats included in this study were swabbed (ear swabs) during routine examination by professional veterinarians either at private veterinary clinics, shelters in the Greater Cairo Area (GCA), and the veterinary hospital of Cairo University. Samples were collected following the consent of the owners. All sampling procedures were "noninvasive no-pain causing procedures". Animal research and reporting of *in vivo* experiments (ARRIVE) guidelines have been completely followed and covered. The legal requirements or guidelines in Egypt for the care and use of animals have been followed, and the institutional animal care and use committee (IACUC) of the Faculty of Veterinary Medicine, Cairo University, Egypt, guidelines have been fully covered. The ethical approval was obtained from the IACUC with approval number (Vet CU131020241033).

Sampling and specimen collection

Samples for this study were collected from September 2022 to September 2023 from 212 pet animals (118 dogs and 94 cats) selected randomly from both sexes of different ages and breeds during their visits to different private veterinary clinics and from shelters in the Greater Cairo Area (GCA), as well as the university veterinary hospital of Cairo University. All animals had no record of previous hospitalization, and all were clinically diagnosed as otitis cases, showing one or more clinical signs suggestive for otitis. Dogs and cats who had previously received topical or systemic treatment with antibiotics or anti-inflammatory drugs 10 days before sample collection were excluded from the study (Levison and Levison, 2009; Baietto *et al.*, 2014). Samples of ear exudate were collected by inserting single-use commercially available sterile cotton-tipped swabs into the horizontal ear canal, and then rolling them out after completing a full 360-degree rotation (Choi *et al*., 2018). Extreme caution was carried out to avoid surface contamination. Such samples were immediately transported to the laboratory in a cool condition (2-8°C) until further processed in the Microbiology Laboratory of the Faculty of Veterinary Medicine, Cairo University.

Isolation and identification

Swabs were inoculated in nutrient broth and were incubated for 24 hr at 37°C. A loopful of culture was streaked on both MacConkey and blood agar plates, and was incubated at 37°C for 24 hr. Pink colonies either with bile precipitation surroundings or mucoid surface lactose fermenter colonies on MacConkey agar were picked up, then sub-cultured on 5% sheep blood agar (Himedia®, India) and incubated to test the hemolytic pattern of the developed colonies. Beta-hemolytic colonies on blood agar were picked up to prepare bacterial microscopic films, and were Gram-stained to ensure the presence of Gram-negative short cocci to coccobacilli. Cultures were then tested for oxidase, urease, triple sugar iron (TSI), indole, methyl red, Vogus Proskauer, and citrate (IMViC) according to methods described by (Kao *et al*., 2016). PCR was used as a molecular confirmation to complete the biochemical identification.

Antimicrobial susceptibility testing of recovered *K. pneumoniae* and *E. coli* isolates

The obtained *K. pneumoniae* and *E. coli* isolates were subjected to antimicrobial susceptibility testing using Muller-Hinton agar by the Kirby-Bauer disc diffusion method. The antibiotic discs used were amoxicillin (AX, 10µg, Himedia®, India), amoxicillin/clavulanic acid (AMC, 20/10µg, Himedia®, India), ceftriaxone (CTR, 30µg, Himedia®, India), ceftazidime (CAZ, 30µg, Himedia®, India), cefoperazone (CPZ, 75µg, Himedia®, India), enrofloxacin (EX, 5µg, Himedia®, India), gentamicin (GEN, 10µg, Himedia®,

India), amikacin (AK, 30µg, Himedia®, India), linezolid (LZ, 30µg, Himedia®, India), tylosin (TL, 15µg, Himedia®, India), lincomycin (LCM, 2µg, Himedia®, India). Diameters of the inhibitory zone obtained around the antibiotic discs were measured after incubation at 37°C for 24 hr and interpreted according to the Clinical and Laboratory Standards Institute (CLSI, 2022).

The ceftriaxone, ceftazidime, and amoxicillin/clavulanic acid discs were placed centered to each other with 30 mm distance from each other to determine the cephalonsporins-clavulanate synergistic effect if they exist as a presumptive phenotypic indication to ESBL production.

- Molecular identification of the obtained isolates and resistance genes detection

DNA extraction

DNA was extracted from all obtained *K. pneumoniae* and *E. coli* isolates using a QIAamp DNA Mini Kit (Qiagen®, Germany) following the manufacturer's instructions.

- Molecular confirmation of the recovered *K. pneumoniae* and *E. coli* isolates

Multiplex PCR was performed using primers (Metabion®, Germany) targeting genes specific for *K. pneumoniae* and *E. coli* listed in Table (1). The amplification step was performed using Emerald Amp GT PCR master mix (Code No. RR310A, Takara®, Japan) according to the following protocol:

- Detection of *bla*TEM and *bla*CTX-M genes of the recovered MDR *K. pneumoniae* and *E. coli* isolates

The *bla*TEM and *bla*CTX-M genes were amplified using the primers (Metabion®, Germany) listed in Table (1). The PCR amplification reaction was prepared and conducted according to the following thermal program illustrated in Tables (2 & 3).

Table (1): Forward and reverse pairs of primers used in the detection of each tested gene

Gene	Primer sequence	Amplified product	Reference
16S rRNA	F: (3`-ATTTGAAGAGGTTGCAAACGAT-5`) R: (3`-TTCACTCTGAAGTTTTCTTGTGTTC-5`)	130 bp	Turton *et al*., 2010
***bla*TEM**	F: (3`-ATCAGCAATAAACCAGC-5`) R: (3`-CCCCGAAGAACGTTTTC-5`)	516 bp	Colom *et al*., 2003
***bla*CTX-M**	F: (3`-ATGTGCAGYACCAGTAARGTKATGGC-5`) R: (3`-TGGGTRAARTARGTSACCAGAAYCAGCGG-5`)	593 bp	Archambault *et al*., 2006

Table 2: The PCR reaction mix components

Component	Volume/reaction
Emerald Amp GT PCR mastermix (2x premix)	12.5µl
PCR grade nuclease-free water	5.5µl
Forward primer (20pmol)	1µl
Reverse primer (20pmol)	1µl
Template DNA	5µl
Total	**25µl**

Table 3: The PCR programs used with each tested gene

Gene	Primary denaturation	Secondary denaturation	Annealing	Extension	Number of cycles	Final extension
16S rRNA	94˚C, 5 min	94˚C, 30 sec	55˚C, 30 sec	72˚C, 30 sec	35	72˚C, 7 min
***bla*TEM**	94˚C, 5 min	94˚C, 30 sec	54˚C, 40 sec	72˚C, 45 sec	35	72˚C, 10 min
***bla*CTX-M**	94˚C, 5 min	94˚C, 30 sec	54˚C, 40 sec	72˚C, 45 sec	35	72˚C, 10 min

RESULTS

In dogs, the total number of collected samples was 118 samples, 70 were males and 48 were females. In relation to the ear type, 76/118 (64.4%) dogs had pendulous ears, while 42/118 (35.6%) had erect or semi-erect ears. 79 were individually owned dogs, and 39 were shelter dogs.

Regarding cats' samples, the total number of collected samples was 94, 55 males and 39 females, 48 were household cats, and 46 were shelter cats. The prevalence of the obtained *K. pneumoniae* and *E. coli* in correlation with habitat and breed has been calculated and represented through Tables (4 and 5).

Table 4: The prevalence of *K. pneumoniae* and *E. coli* in correlation with habitat

Bacteria	Cat		Dog	
	Household	Shelter	Household	Shelter
K. pneumoniae	11	22	13	17
E. coli	8	9	9	12

Table 5: The prevalences of *K. pneumoniae* and *E. coli* in correlation with dogs' breeds

Breed	Bacteria	
	K. pneumoniae	***E. coli***
Yorkshire	0	0
Shih Tzu	0	0
Poodle	0	0
Pug	0	1
French Bulldog	1	0
Rottweiler	2	2
Griffon	2	3
Beagle	1	1
German Shepherd	2	2
Mongrel	10	4
Cocker Spaniel	6	4
Retrievers	6	4

Among the 118 dogs and 94 cats, isolation rates of *K. pneumoniae* in the otitis cases were 18% in both dogs and cats, while those for *E. coli* were 12.4% and 9.6%, respectively. The highest rate of *E. coli* resistance, in both dog and cat isolates, was found in linezolid, tylosin, and lincomycin (100%), followed by amoxicillin (71%-88%) and amoxicillin/clavulanic acid (62%-71%), from dogs and cats, respectively. A total of 17% of cat isolates were resistant to ceftriaxone, gentamicin, and amikacin, while the resistance rate for dog isolates was 62%, 14%, and 9%, respectively. The rates of resistance to ceftazidime, cefoperazone, and enrofloxacin were 29%-57%, 47%-62%, and 59%-24% of cat and dog isolates, respectively Table (6) and Figure (1).

Regarding the *K. pneumoniae* resistance rate, in both dogs and cats' isolates, was found in linezolid, tylosin, and lincomycin (100%), while in amoxicillin and amoxicillin/ clavulanic acid was also (100%) in dog isolates only and (91%-88%) in cat isolates, respectively. Followed by cefoperazone, ceftazidime, ceftazidime, ceftriaxone, and enrofloxacin as (27%-54%), (33%-48%), (33%-40%), and (33%-33%) from dogs and cats, respectively. A total of 9% of cat isolates were resistant to gentamicin, and 6% were resistant to amikacin, while the resistance rate for the same dog isolates was 17%, and 10%, respectively Table (6) and Figure (1).

PCR screening of isolated bacteria revealed all recovered *K. pneumoniae* and *E. coli* harbored ESBL genes, *bla*TEM, and *bla*CTX-M. Detection, virulence, and resistance genes of screened bacterial isolates are detailed in Table (6).

Table 6: A detailed antibiotic susceptibility collective profile of the obtained isolates illustrated as numbers and percent

Drug	*K. pneumoniae*						*E. coli*					
	Dog			Cat			Dog			Cat		
	S	I	R	S	I	R	S	I	R	S	I	R
AX	0 (0%)	0 (0%)	30 (100%)	2 (6%)	1 (3%)	30 (91%)	6 (29%)	0 (0%)	15 (71%)	2 (12%)	0 (0%)	15 (88%)
AMC	0 (0%)	0 (0%)	30 (100%)	2 (6%)	2 (6%)	29 (88%)	8 (38%)	0 (0%)	13 (62%)	5 (29%)	0 (0%)	12 (71%)
CTR	15 (50%)	5 (17%)	10 (33%)	9 (27%)	11 (33%)	13 (40%)	6 (29%)	2 (9%)	13 (62%)	12 (71%)	2 (12%)	3 (17%)
CAZ	15 (50%)	5 (17%)	10 (33%)	15 (46%)	2 (6%)	16 (48%)	4 (19%)	5 (24%)	12 (57%)	10 (59%)	2 (12%)	5 (29%)
CPZ	15 (50%)	7 (23%)	8 (27%)	13 (40%)	2 (6%)	18 (54%)	6 (29%)	2 (9%)	13 (62%)	5 (29%)	4 (24%)	8 (47%)
EX	20 (67%)	0 (0%)	10 (33%)	15 (46%)	7 (21%)	11 (33%)	16 (76%)	0 (0%)	5 (24%)	7 (41%)	0 (0%)	10 (59%)
GEN	23 (76%)	2 (7%)	5 (17%)	29 (88%)	1 (3%)	3 (9%)	18 (86%)	0 (0%)	3 (14%)	14 (83%)	0 (0%)	3 (17%)
AK	24 (80%)	3 (10%)	3 (10%)	30 (91%)	1 (3%)	2 (6%)	19 (91%)	0 (0%)	2 (9%)	14 (83%)	0 (0%)	3 (17%)
LZ	0 (0%)	0 (0%)	30 (100%)	0 (0%)	0 (0%)	33 (100%)	0 (0%)	0 (0%)	21 (100%)	0 (0%)	0 (0%)	17 (100%)
TL	0 (0%)	0 (0%)	30 (100%)	0 (0%)	0 (0%)	33 (100%)	0 (0%)	0 (0%)	21 (100%)	0 (0%)	0 (0%)	17 (100%)
LCM	0 (0%)	0 (0%)	30 (100%)	0 (0%)	0 (0%)	33 (100)	0 (0%)	0 (0%)	21 (100%)	0 (0%)	0 (0%)	17 (100%)

S; sensitive, I; intermediate, and R; resistant. AX; amoxicillin, AMC; amoxicillin/clavulanic acid, CTR; ceftriaxone, CAZ; ceftazidime, CPZ; cefoperazone, EX; enrofloxacin, GEN; gentamicin, AK; amikacin, L2; linezolid, TL; tylosin, and LZ; lincomycin.

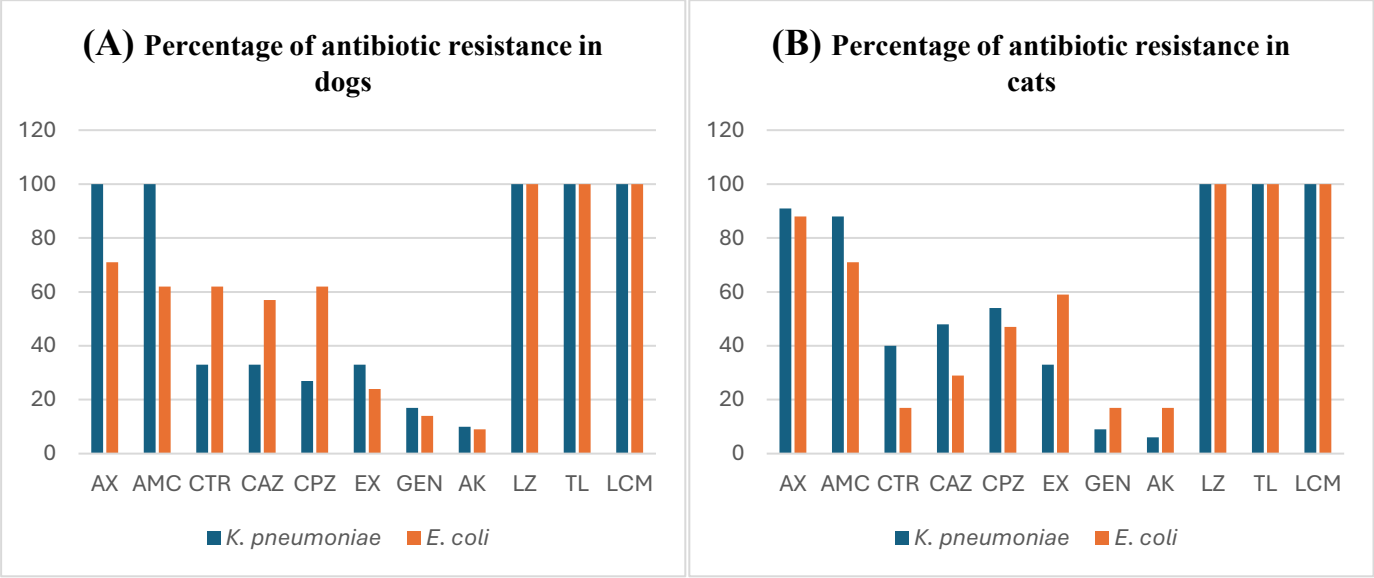

Figure 1: Clustered column charts illustrating the percentage of antibiotic resistance in dogs (A) and cats (B).

DISCUSSION

To control AMR, prevention and intervention strategies must consider the One Health concept, with proper consideration of animal-human companionship that plays a crucial role in this approach. Surveillance is a crucial part of developing an evidence-based plan. Numerous sectors are included in the complicated AMR surveillance study, and methodological considerations are essential for interpreting the surveillance data further, including choosing a credible sentinel bacterial host with AMR

characteristics. Thus, the One Health components require scientific evidence. Since human medications can be used on companion animals, ESBL-producing Enterobacterales are more likely to use reserved antimicrobial drugs for human use.

Pet cats and dogs could be a possible source of disease transmission to human beings, even though they may offer their owners several psychosocial benefits (Bhat, 2021). Regarding antibiotic resistance, the current study examined the ear carriage of ESBL-MDR-*K. pneumoniae* and *E. coli* in pets, including dogs and cats. Although some articles mentioned the occurrence of ESBL *K. pneumoniae* and *E. coli* from urinary tract infections (Facchin *et al*., 2025), lower GIT (van den Bunt *et al*., 2020), and upper respiratory tract (Yoon *et al.,* 2024), there are, as far as we are aware, no prior studies on ESBL-MDR *K. pneumoniae* and *E. coli* causing otitis in animals to compare the results with. Therefore, regardless of the source of the specimen, the comparison concentrated on the presence of ESBL-MDR *K. pneumoniae* and *E. coli* in dogs and cats.

According to the findings of the current study, *K. pneumoniae* was more common in instances of otitis in both dogs and cats (18%) than *E. coli* (12.4% and 9.6%, respectively). These results contradict the findings of (Elmeslemany and Younis, 2023), which revealed that *E. coli* was the most common species among the dogs and cats under investigation. However, they are consistent with the findings of (Woerde *et al*., 2023), which were derived from ducks as the sole study that coincided with the current work results about the greater *K. pneumoniae* occurrence rate. Remarkably, the current findings show that ESBL-KP and ESBL-EC are highly prevalent in the ear canals of dogs and cats analyzed (17.1% and 10.4%, respectively). These values are comparable and higher than that obtained by (Poirel *et al*., 2013; Huber *et al*., 2013; Liao *et al*., 2013; So *et al*., 2012; Sun *et al*., 2010; O'keefe *et al*., 2010; Rumi *et al*., 2019; Haenni *et al*., 2014; Zogg *et al*., 2018; Karkoba *et al*., 2019; van den Bunt *et al*., 2020; Pepin-Puget *et al*., 2020; Carvalho *et al*., 2020; Ewers *et al*., 2014) who studied ESBL-KP and/or ESBL-EC obtained from wide range of infections in dogs and/or cats. Also, household and shelter animals were involved in the current work, and regardless of the level and nature of care given to each of them based on the habitat, both household and shelter animals were found to be a possible source of ESBL-MDR *K. pneumoniae* and *E. coli.* But it was noticed that the infection rate was higher in both dogs' and cats' cases obtained from shelters than those obtained from household habitats, reflecting the effect of outdoor exposure and multi-animal living environment on increasing the possibilities of infection spread.

Because pet owners frequently come, both directly and indirectly, into close contact with their pets (Stull *et al.*, 2012; Stull *et al*., 2013), the high ear carriage rates of ESBL-KP and ESBL-EC in dogs and cats pose a possible source of infection transmission to humans (Stull *et al.*, 2014). These organisms can enter the human body through skin abrasions or hand contamination causing specific clinical manifestations according to the causative agent. It has been previously presented that serious fungal, bacterial, and viral pathogens may find their way to establish human infections employing animal-human companionship, such as dermatophytes (Hassanien *et al.*, 2021), rhodotorulae (Aboul-Ella *et al.*, 2025), staphylococci (Haag *et al*., 2019; Hamdy *et al.,* 2025), enterococci (Hammerum, 2012), *E. coli*, *K. pneumoniae*, *P. mirabilis*, *P. aeruginosa* (Hamdy *et al.,* 2025), and Rabies (Soliman *et al*., 2024). Establishment of a microbial infection disturbs the microbiome community and the immune balance and leads to immune dysregulation and susceptibility to infection (Kogut *et al.,* 2020). Secreta and excreta of dogs and cats may provide a possible source of contamination to households and a source

of infection to humans (Tan, 1997), and ESBL-KP and ESBL-EC may survive on contaminated objects for many days (Ncir *et al*., 2024).

Given the information above, pets—particularly dogs—may serve as a significant reservoir for ESBL-KP and ESBL-EC in the community or several nosocomial outbreaks. Additionally, all phenotypically identified ESBL-producing isolates through the current work were MDR and had both the *bla*CTX-M and *bla*TEM genes. It is worth mentioning that this is the first report of ear canal colonization of ESBL-KP and ESBL-EC possessing the *bla*CTX-M and *bla*TEM genes among pet dogs and cats in the community. This should be considered in the progression and evolution of community-acquired ESBL-KP and ESBL-EC infections (Chen *et al*., 2021).

*Bla*CTX-M was the most prevalent ESBL gene and is the most common ESBL gene reported in *E. coli* isolates from dogs and cats worldwide (Salgado-Caxito *et al*., 2021). The prevalence of various ESBL genes in bacteria from companion animals may vary geographically, although the number of isolates in these studies is low. The *bla*TEM gene family was the most common in a series of studies conducted in Brazil (Sfaciotte *et al*., 2021), France (Dupouy *et al*., 2019), and New Zealand (Karkaba *et al*., 2019). A study performed in 2011 that characterized ESBL genes from 54 *E. coli* isolates from companion animals in the USA revealed 78% of isolates carried the *bla*CTX-M gene (Shaheen *et al*., 2011). Concerningly, all isolates in our study possessed both genes, *bla*CTX-M and *bla*TEM, that could contribute to beta-lactam resistance.

Though numerous investigations that characterized ESBL-KP and ESBL-EC isolates from animals (Liu *et al*., 2021) and humans (Saleem *et al*., 2017) instances failed to discover either *bla*CTX-M or *bla*TEM genes, although the *bla*CTX-M and *bla*TEM genes have been previously detected in hospital-associated ESBL-EC infections from dogs and cats (Mitu *et al*., 2019). In a sole study, ESBL-KP and ESBL-EC isolates obtained from the feces of healthy dogs were found to contain the *bla*CTX-M and *bla*TEM genes (Belas *et al*., 2021). Serious nosocomial infections in hospitals are typically caused by ESBL-KP and ESBL-EC strains that possess the *bla*CTX-M and *bla*TEM genes (Akenten *et al*., 2023). The present results demonstrate that ESBL-KP and ESBL-EC may have a zoonotic route that extends outside medical facilities.

CONCLUSION

The recent study showed the ear carriage of multidrug-resistant and ESBL-producing bacteria among pet dogs and cats with otitis, emphasizing the importance of enhanced global surveillance for pet animals-related antimicrobial resistance and the need for more focus on companion animals' roles in AMR epidemiology, particularly given that the antimicrobials utilized are also used in human medicine.

FUTURE RESEARCH

To ascertain if the phenotypic and genotypic traits of ESBL-producing bacteria from healthy companion animals differ from those from diseased companion animals, more research is required. Additional studies are also needed to identify risk factors for infection and the outcomes of infection, as companion animals require a better understanding of prevalence, risk factors, consequences, and mechanisms of resistance gene acquisition

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Conflict of interest

The authors declare that they have no conflict of interest

Authors Contributions

The study was performed by Mayson Hamdy, designed and supervised by Sherif Marouf, Soliman Mohamed, and Haitham Farghali, while Hassan Aboul-Ella contributed to implementation of the work, analysis of the results, and writing the manuscript.

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نظرة ثاقبة على بكتيريا الكلبسيلة الرئوية والإشريكية القولونية متعددة المقاومة للمضادات الحيوية والمنتجة لإنزيم البيتالاكتاميز المثبط بين الحيوانات الأليفة التي تعاني من التهاب الأذن

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أُجريت هذه الدراسة لدراسة استعمار قناة الأذن ببكتيريا الكلبسيلة الرئوية والإشريكية القولونية متعددة المقاومة للمضادات الحيوية والمنتجة لإنزيم البيتالاكتاميز المثبط (ESBL-KP وESBL-EC) لدى الحيوانات الأليفة التي شُخِّصت سريريًا على أنها حالات التهاب أذن معدي. كما وُجدت علاقة ارتباط بين معدل العزل والعديد من الظروف المهيئة لحدوث المرض، مثل العمر ومكان وطبيعة المعيشة وسلالة الحالات المدرجة بالدراسة. تم الحصول على مسحات الأذن من ١١٨ و٩٤ كلبًا وقطا أليفًا، على التوالي، ليس لديهم تاريخ مرضي سابق معروف. تم إثراء جميع العينات على أجار ماكونكي الانتقائي وأجار دم الأغنام بنسبة ٥٪ لعزل انواع البكتيريا المعوية موضع الاهتمام. تم التعرف على الكلبسيلة الرئوية والإشريكية القولونية *K. pneumoniae* و *E. coli* بشكل أكبر من خلال التقنيات البيوكيميائية والجزيئية. تم تحديد أنماط مقاومة مضادات الميكروبات للمعزولات المُستخرَجة عليها بواسطة طريقة كيربي باور، وتم فحص السلالات المقاومة للأدوية المتعددة بواسطة تفاعل البلمرة المتسلسل (PCR) للكشف عن جيني المقاومة *bla*CTX-M و *bla*TEM . بلغت معدلات انتشار ESBL-KP في الأذن بين الكلاب والقطط الأليفة ١٧.٨٪ و١٨% على التوالي، بينما بلغت معدلات انتشار ESBL-EC ١٢.٤٪ و٩.٦٪ على التوالي. . لم تُظهر أيٌّ من معزولات سلالات الكلبسيلة الرئوية والإشريكية القولونية *K. pneumoniae* و *E. coli* حساسيةً لليفوفلوكساسين والتيلوسين واللينكومايسين. ومع ذلك، وجدت بكتيريا الكلبسيلة الرئوية والإشريكية القولونية K. pneumoniae وE. coli المقاومة للسيفترياكسون والسيفتازيديم والسيفوبيرازون في العينات المفحوصة بمعدلات ١٧.٦٪ و١٩.٤٪ و٢١.٢٪ على التوالي. علاوة على ذلك، أظهرت جميع معزولات *K. pneumoniae* و *E. coli* مقاومة للأدوية المتعددة. في حين تم اكتشاف جينات *bla*CTX-M و*bla*TEM في جميع معزولات متعددة المقاومة من الكلاب والقطط. ويسلط ظهور جينات *bla*CTX-M و*bla*TEM الضوء على دور الحيوانات الأليفة كمصدر محتمل لانتقال مثل هذه مسببات الأمراض إلى البشر.