Multidrug resistance of *Escherichia coli* isolated from the urinary bladder of dogs and cats with suspected urinary tract infections

Dawid Jańczak[1,A-D]✉, Piotr Górecki[1,B-C], Rafał Stryjek[2,C,E-F], Aleksandra Zasada[3,C,E-F]

[1] ANIMALLAB Veterinary Laboratory, Warsaw, Poland
[2] Institute of Psychology, Polish Academy of Sciences, Warsaw, Poland
[3] Department of Sera and Vaccines Evaluation, National Institute of Public Health NIH – National Research Institute, Warsaw, Poland

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▌Abstract

Introduction and Objective. Escherichia coli is one of the most common bacteria isolated from urine samples collected from dogs and cats with urinary tract infection (UTI). Uncomplicated UTIs in dogs and cats can be treated with short courses of first-line antimicrobial drugs, e.g. amoxicillin, amoxicillin with clavulanic acid, or trimethoprim/sulfonamide. Recurrent or complicated UTIs often require long-term treatment with broad-spectrum antibiotics. However, the choice of drug should be based on antimicrobial susceptibility.

Materials and method. Between March – September 2022, E. coli isolates cultured from the urine of 66 dogs and 41 cats with UTI symptoms were tested for antimicrobial resistance by using Minimum Inhibitory Concentration (MIC). Antimicrobial susceptibility was tested for ampicillin, ampicillin/sulbactam, cefazolin, cefuroxime, aztreonam, gentamycin, amikacin, colistin, trimethoprim/sulfamethoxazole, ciprofloxacin, chloramphenicol and tetracycline.

Results. The highest prevalence of resistance was documented for ampicillin (68% in dogs, 100% in cats) and ampicillin with sulbactam (59% in dogs, 54% in cats). The most common antimicrobial resistance patterns of E. coli were ampicillin alone (12 isolates, 29.3% in cats) and beta-lactams, including aztreonam (14 isolates, 21.2% in dogs).

Conclusions. High resistance to aztreonam (61% and 32% of isolates from dogs and cats, respectively), other beta-lactams, and fluoroquinolones should cause be alarm due to zoonotic potential and cross-transmission of antimicrobial-resistant microorganisms between animals and humans.

▌Key words

Escherichia coli, urinary tract infection, public health, multidrug resistance, dogs, cats

INTRODUCTION

Urinary tract infections (UTIs) are one of the most frequently diagnosed diseases in dogs and cats [1, 2, 3]. Veterinary medicine distinguishes complicated and uncomplicated types of UTIs [4]. Uncomplicated UTI can occasionally be diagnosed in healthy animals with normal anatomy and physiology of the urinary tract which can be resolved after a few days of treatment with first-line antimicrobials [1, 2]. However, many co-occurring conditions, such as hyperadrenocorticism, hyperthyroidism, chronic kidney disease (CKD), diabetes mellitus, neoplasia, obesity, urolithiasis infectious diseases, and urinary tract anatomical or physiological disorders, incur a risk of complicated and recurrent UTI which is difficult to treat with broad-spectrum antimicrobials [1, 2, 3, 4, 5].

Bacterial UTI occur less frequently among cats (1–2%) than in dogs (~ 14%) in their lifetime and depends on the age of the animal [1, 6]. However, distinguishing bacterial cystitis, idiopathic cystitis, clinical or subclinical bacteriuria is challenging [1].

Many bacterial isolates responsible for UTI in companion animals are an important issue for public health [7, 8]. The European Centre for Disease Prevention and Control (CDPC) mentions the spread and cross-transmission of antimicrobial-resistant microorganisms between animals, between humans, and between humans and animals and the environment as one of the two major drivers for antimicrobial resistance [9]. Multidrug-resistant (MDR) bacteria with zoonotic potential are often isolated from dogs and cats with UTI, mainly *Escherichia coli*, *Klebsiella* spp., *Staphylococcus* spp., *Enterococcus* spp., *Proteus mirabilis,* and *Pseudomonas* spp. [5, 10]. Complete diagnosis of bacterial UTI includes urinalysis with sediment examination and culture. Urine collection by cystocentesis is highly recommended to prevent sample contamination [2, 11]. *Escherichia coli* is the most common Gram-negative bacteria isolated from urine samples of canines (39–58.8%) and felines (38.8–59.5%) with suspected UTI [1, 5, 6, 12, 13, 14, 15]. Selection of the most effective antimicrobial drug should be based on the antimicrobial susceptibility test, and the treatment can be started pending the susceptibility results by administering first-line antimicrobials including amoxicillin or trimethoprim/sulfonamide [1, 2]. Because of the potential zoonotic risk, the topic of this paper is the antimicrobial resistance to agents used in human and veterinary medicine among *Escherichia coli* isolates from dogs and cats urine samples was

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MATERIALS AND METHOD

A source of bacterial isolates. Between March – September 2022, 107 *Escherichia coli* isolates were cultured from urine samples collected by cystocentesis from cats (n=41) and dogs (n=66) with pyuria. Pyuria was confirmed by microscopy when there were more than 5 leucocytes in a field-of-view under 400× total magnification. For samples with massive pyuria, >50 leucocytes in the field-of-view cytology, Giemsa staining was made (Fig. 1). All cultured isolates were >1000 CFU/mL. No other bacteria species were cultured. Since the urine samples were sent by veterinarians as a part of the laboratory service tests, an Ethic Commission Agreement was not required.
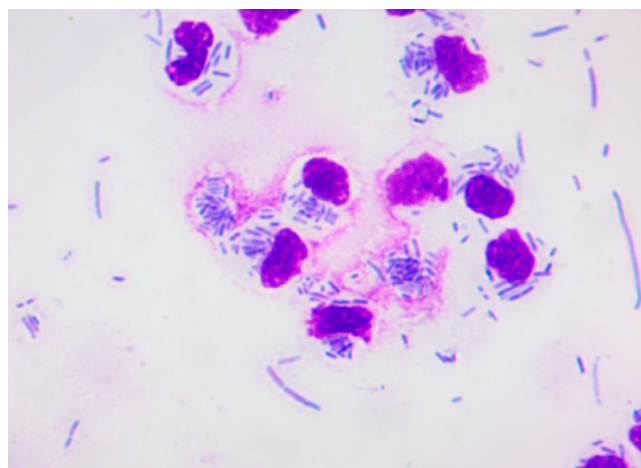
Figure 1. Cytology of urine sediment with visible leucocytes containing rod-shaped bacteria. Objective magnification 100×

Urine samples were inoculated onto 5% defibrinated sheep blood agar, MacConkey's agar plates (Graso Biotech), and UriSelect™4 Agar (Bio-Rad). The plates were incubated in aerobical conditions at 35±2 °C for 24, 48 and 72 hours. Bacteria were identified based on colony growth morphology, Gram staining, catalase and oxidase tests, and biochemical properties by using commercial test GN24 (DIAGNOSTICS s.r.o., Slovakia) for glucose fermenting / nonfermenting Gram negative bacteria.

Among the 41 cats, 8 were male, and 33 were female. The oldest cat was 18-years-old, and the youngest – eight months old (median age = 8 years and 4 months). Among the 66 dogs, 16 were male and 50 were female. The oldest dog was 18-years-old, and the youngest – 6 months old (median age = 11 years).

Minimum Inhibitory Concentration (MIC). The lowest concentration of the antimicrobial agent which visibly inhibits bacterial growth was tested with commercial kit MIC GN 1 (DIAGNOSTICS s.r.o., Slovakia) including antimicrobial agents: Ampicillin (AMP), Ampicillin/Sulbactam (AMS), Cefazolin (CFZ), Cefuroxime (CXM), Aztreonam (AZT), Gentamycin (GEN), Amikacin (AMK), Colistin (COL), Trimethoprim/Sulfamethoxazole (T/S), Ciprofloxacin (CIP), Chloramphenicol (CMP) and Tetracycline (TET) was used. Bacterial suspension was performed according to the manufacturer's instructions: 60 µl well homogenized in sterile saline suspension of turbidity of 0.5 McF was added to 12 mL of suspension media. Next, 100 µL of bacterial suspension was inoculated into each microplate well. Test results were read visually and compared to the positive control after 20 hours of incubation at 35 °C.

Antimicrobial susceptibility for *E. coli* was determined based on the MIC breakpoints of the veterinary VET08, VET01S and human M100-specific CLSI guidelines if the MIC breakpoints were not available in the veterinary CLSI guidelines [16, 17, 18]. The MIC breakpoints for ciprofloxacin, cefazoline, cefuroxime, and colistin were based on available publications [8, 19, 20]. Classification of the prevalence of antimicrobial resistance (AMR) of *E. coli* was as follows: rare – <0.1%, very low – 0.1–1.0%, low – > 1.0–10.0%, moderate – > 10.0–20.0%, high – > 20.0–50.0%, very high – >50.0–70.0%, and extremely high – > 70.0%. The European Union Summary Report on Antimicrobial Resistance in zoonotic and indicator bacteria from humans, animals, and food in 2018–2019 was used [21]. MIC 50 and MIC 90 values and the most common antimicrobial resistance patterns were also determined.

Statistical analysis. Non-parametric tests were employed to account for non-normal distributions and unequal sample sizes. A one-sided Spearman correlation was utilized to assess the correlation between age and drug resistance, and the Mann-Whitney U test employed for pairwise comparisons between genders and species. Statistical analysis was performed using IBM SPSS Statistics software (IBM Corp., 2019).

RESULTS

Antimicrobial resistance of Escherichia coli in dogs. *Escherichia coli* isolates had a very high prevalence of resistance to ampicillin (68%), ampicillin/sulbactam (59%), cefazoline (52%), cefuroxime (64%), aztreonam (61%) and amikacin (53%). They also had a high prevalence for trimethoprim/sulfamethoxazole (23%), tetracycline (36%), and 47%, respectively, for ciprofloxacin and chloramphenicol. Moderate resistance was determined for gentamycin (14%) and low for colistin (8%) (Tab 1). The often-occurring AMR patterns in *Escherichia coli* isolates were isolates' resistance to beta-lactams, including ampicillin, ampicillin/sulbactam, cefazoline, cefuroxime and aztreonam (14 isolates, 21.21%), ampicillin alone (4 isolates, 6.06%), beta-lactams and ciprofloxacin (5 isolates, 7.57%), and beta-lactams with aminoglycosides, trimethoprim/sulfamethoxazole, ciprofloxacin, chloramphenicol, and tetracycline (5 isolates, 7.57%) (Tab 2).

Antimicrobial resistance of Escherichia coli in cats. *Escherichia coli* isolates had an extremely high prevalence of resistance to ampicillin (100%), very high to ampicillin/sulbactam (54%), and high to amikacin (39%) and aztreonam (32%). A 37% prevalence was determined for cefazoline, cefuroxime, ciprofloxacin, and chloramphenicol. Moderate resistance was determined for trimethoprim/sulfamethoxazole (17%), and low for colistin (10%) and gentamycin (10%) (Tab 3). The often-occurring AMR pattern in *Escherichia coli* isolates were resistance to beta-lactams, including ampicillin, ampicillin/sulbactam, cefazoline, cefuroxime and aztreonam (5 isolates, 12.2%), ampicillin alone (12 isolates, 29.3%), ampicillin, ampicillin/sulbactam and ciprofloxacin (5 isolates, 12.2%) (Tab. 4).

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Table 1. Antimicrobial resistance in *Escherichia coli* isolated from dog's urine samples

Dogs	MIC (µg/mL)														%	%	(µg/mL)	
Antibiotics	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	>128	S	R	MIC$_{50}$	MIC$_{90}$
AMP						7	9	4	1	0	0	2	2	41	32	68	64	>128
AMS						4	12	8	3	3	6	1	6	23	41	59	32	>128
CFZ			3	0	0	0	3	17	7	2	34				48	52	>16	>16
CXM						3	0	13	8	6	2	4	4	26	36	64	32	>64
AZT			22	2	2	0	0	0	0	0	40				39	61	>16	>16
GEN					3	4	25	25	0	0	0	3	6		86	14	2	32
AMK						4	1	15	11	0	4	28	2	1	47	53	16	32
COL			1	0	9	21	30	5	0	0	0				92	8	2	2
T/S	3	2	16	11	11	3	5	0	15						77	23	0.5	>4
CIP		25	9	1	0	0	8	6	4	13					53	47	0.25	>8
CMP				2	1	0	2	5	25	0	0	31			53	47	8	>32
TET					3	1	4	34	0	0	20	4	0		64	36	4	32

Table 2. The most common antimicrobial resistance patterns of *Escherichia coli* isolated from dog's urine samples.

Antimicrobial resistance patterns	No. of antimicrobial classes in pattern	n	(%)
AMP	1	4	6.1
AMP-AMS-CFZ-CXM-AZT	1	14	21.2
AMP-AMS-CFZ-CXM-AZT-CIP	2	4	6.1
CIP	1	3	4.5
CMP	1	2	3.0
AMP-T/S-CIP-TET	4	2	3.0
AMP-AMS-CFZ-CXM-AZT-TET	2	2	3.0
AMP-AMS-CFZ-CXM-AZT-GEN-AMK-T/S-CIP-CMP-TET	6	2	3.0
AMP-AMS-CFZ-CIP	2	2	3.0
TET	1	1	1.5
T/S-CIP-CMP-TET	4	1	1.5
T/S-CIP	2	1	1.5
T/S	1	1	1.5
AMP-GEN-T/S-CIP-TET	5	1	1.5
AMP-AMS-CFZ-CXM-AZT-T/S-CIP-TET	4	1	1.5
AMP-AMS-CFZ-CXM-AZT-T/S-CIP-CMP-TET	5	1	1.5
AMP-AMS-CFZ-CXM-AZT-T/S-CIP	3	1	1.5
AMP-AMS-CFZ-CXM-AZT-GEN-COL-T/S-TET	5	1	1.5
AMP-AMS-CFZ-CXM-AZT-GEN-AMK-T/S-CIP	4	1	1.5
AMP-AMS-CFZ-CXM-AZT-GEN-AMK-CIP-CMP-TET	5	1	1.5
AMP-AMS-CFZ-CXM-AZT-COL-T/S-CMP-TET	5	1	1.5
AMP-AMS-CFZ-CXM-AZT-COL-CMP-TET	4	1	1.5
AMP-AMS-CFZ-CXM-AZT-COL	2	1	1.5
AMP-AMS-CFZ-CXM-AZT-CMP	2	1	1.5
AMP-AMS-CFZ-CXM-AZT-CIP-TET	3	1	1.5
AMP-AMS-CFZ-CXM-AZT-CIP-CMP-TET	4	1	1.5
AMP-AMS-CFZ-CXM-AZT-AMK-T/S-TET	4	1	1.5
AMP-AMS-CFZ-CXM-AZT-AMK	2	1	1.5
AMP-AMS-CFZ-COL-T/S-CIP-CMP	5	1	1.5
AKN-CIP	2	1	1.5
Susceptible		**10**	**15.2**

Correlation between age, gender and antimicrobial resistance in dogs. A moderate positive correlation was observed between dogs' age and drug resistance ($r\_s(64) = 0.476$; $p < 0.001$). Subgroup analyses based on gender revealed a moderate positive correlation in females ($r\_s(48) = 0.511$; $p < 0.001$). However, for males, the correlation was not statistically significant, although it approached a level of marginal significance ($r\_s(14) = 0.368$; $p = 0.081$). No significant differences in drug resistance were found between genders (Mann-Whitney U test, $U = 399$; $p > 0.05$).

Correlation between age, gender and antimicrobial resistance in cats. No significant positive correlation was found between a cat's age and drug resistance, reaching a level of marginal significance ($r\_s(39) = 0.217$; $p = 0.087$). Correlations based on gender subgroups were also not significant for females ($r\_s(31) = 0.27$; $p = 0.064$) and males ($r\_s(6) = 0.15$; $p > 0.05$). Additionally, no significant differences in drug resistance were observed between genders (Mann-Whitney U test, $U = 92$; $p > 0.05$).

Interspecies Comparison. No significant differences in drug resistance were noted between cats and dogs (Mann-Whitney U test, $U = 1126$; $p > 0.05$).

DISCUSSION

Escherichia coli is the most frequent bacteria causing UTI in humans and animals. A multicentre analysis conducted on the Polish population in 2013 found that *E. coli* was responsible for 71.4% of all UTIs [22]. In The Netherlands, among isolated uropathogens from women with symptoms of uncomplicated UTI, *E. coli* was the most prevalent (83%) [23]. In Italy, *E. coli* was the most common pathogen isolated from urine samples of dogs (38.7%) and cats (34.5%). In the same study, mixed bacterial infections were significantly low, 6.3% and 5% in dogs and cats, respectively [24]. Between 2008 – 2010, a pan-European antimicrobial surveillance initiative called ComPath evaluated a minimum inhibitory concentration for uropathogens cultured from 616 urine samples of dogs and cats with UTI symptoms. Similarly, in the current study, the most common isolated pathogen

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Table 3. Antimicrobial resistance in *Escherichia coli* isolated from cats' urine sample

Cats	MIC (µg/mL)														%	%	(µg/mL)	
Antibiotics	0.032	0.064	0.125	0.25	0.5	1	2	4	8	16	32	64	128	>128	S	R	MIC50	MIC90
AMP						0	5	5	2	2	2	0	5	20	0	100	128	>128
AMS						5	7	4	3	8	4	0	2	8	46	54	16	>128
CFZ			1	2	1	0	2	16	3	1	15				63	37	8	>16
CXM						3	0	14	9	0	1	3	11	0	63	37	4	64
AZT			22	1	4	1	0	0	0	2	11				68	32	0.25	>16
GEN				3	3	18	13	0	0	0	0	4			90	10	2	>32
AMK					0	1	13	11	0	0	0	14	2		61	39	4	64
COL			0	0	7	12	17	1	3	0	0				90	10	2	2
T/S	1	6	17	5	1	3	1	0	7						83	17	2	8
CIP		17	5	4	0	0	0	0	10	5					63	37	2	8
CMP				0	0	0	9	2	15	0	0	15			63	37	8	>32
TET					2	1	7	20	0	0	11	0	0		73	27	4	32

Table 4. The most common antimicrobial resistance patterns of *Escherichia coli* isolated from cats' urine samples

Antimicrobial resistance patterns	No. of antimicrobial classes in pattern	n	(%)
AMP	1	12	29.3
AMP-AMS-CFZ-CXM-AZT	1	5	12.2
AMP-AMS-CIP	2	4	9.8
AMP-AMS-TET	2	3	7.3
AMP-AMS	1	2	4.9
AMP-COL-CIP	3	2	4.9
AMP-AMS-CFZ-CXM	1	2	4.9
AMP-AMS-CFZ-CXM-AZT-AMK	2	1	2.4
AMP-AMS-CFZ-CXM-AZT-CIP	2	1	2.4
AMP-AMS-CFZ-CXM-AZT-CMP-TET	3	1	2.4
AMP-AMS-CFZ-CXM-AZT-COL-T/S-CIP-CMP-TET	6	1	2.4
AMP-AMS-CFZ-CXM-AZT-GEN-AMK-T/S-CIP-TET	5	1	2.4
AMP-AMS-CFZ-CXM-AZT-T/S	2	1	2.4
AMP-AMS-CFZ-CXM-AZT-T/S-CIP-CMP-TET	5	1	2.4
AMP-AMS-CFZ-CXM-AZT-T/S-CIP-TET	4	1	2.4
AMP-AMS-T/S-CIP	3	1	2.4
AMP-AMS-T/S-CIP-CMP	4	1	2.4
AMP-COL-TET	3	1	2.4
Susceptible	**0**	**0**	**0**

was *Escherichia coli,* 59.8% and 46.7% in dogs and cats, respectively [25]. A retrospective study between 2017 – 2021 carried out in Portugal also revealed *E. coli* as the most commonly isolated pathogen (44.5%) from urine samples of companion animals [6]. In Poland between 2007 – 2013, Rzewuska et al. (2015) collected 244 isolates of *E. coli* from dogs and cats urine to determine their susceptibility [26].

The number of multidrug-resistant bacteria is increasing, especially in animals that have already been treated with antimicrobials [27]. Inadequate, often empirical choice of antibiotic, failed or shortened administration by pet owners may lead to antimicrobial resistance for common antimicrobial agents administered orally as a first-line treatment. Secondly, public health will be negatively impacted due to antibiotic resistance [2, 6, 28].

The presented study documented a high resistance level to commonly administered antimicrobials, such as ampicillin and other beta-lactams, including 1st and 2nd generations of cephalosporins and aztreonam. A high prevalence of resistance was also documented for ciprofloxacin and chloramphenicol.

In veterinary medicine, amoxicillin alone or amoxicillin with clavulanic acid is a reasonable drug of choice for bacterial cystitis in short-term treatment. Trimethoprim with sulfonamides can also be used as first-line treatment [2].

In human medicine, beta-lactams and fluoroquinolones are considered as second-line treatment, in contrast to nitrofurantoin, trimethoprim-sulfamethoxazole or fosfomycin, which are recommended as the drugs of choice in first-line treatment. A short fluoroquinolone course is recommended in patients with uncomplicated pyelonephritis who do not require hospitalization [29].

Davis et al. (2022), based on 329 positive bacterial culture growths from a county hospital on the US – Mexico border, documented a high resistance for trimethoprim-sulfamethoxazole, tetracycline, ciprofloxacin, levofloxacin, and cephalexin. At the same time, the lowest resistance was noted for amoxicillin with clavulanic acid, cefdinir, cefuroxime, and nitrofurantoin. Furthermore, nitrofurantoin was the most commonly used antibiotic in the treatment of UTI or cystitis [30]. Haddad et al. (2020) indicate that considering the local pathogen resistance patterns, antibiotics should only be used in cases of symptomatic UTI [31]. The empirical treatment of UTI is becoming more challenging because of increasing antimicrobial resistance.

Due to their broad spectrum and easy administration, the frequent use and prescription of fluoroquinolones in human and veterinary medicine leads to significant resistance [32, 33]. Among fluoroquinolones, enrofloxacin is commonly used in veterinary medicine. A 3-day course of oral enrofloxacin in a dose of 20 mg/kg once a day is effective for treating uncomplicated UTI in dogs because of the additive effectiveness of enrofloxacin and its primary metabolite, which is ciprofloxacin [34]. In the current study, resistance to ciprofloxacin revealed 47% and 37% in dogs and cats, respectively. In the study by Feßler et al., resistance to ciprofloxacin was detected in 23 (39%) among 59 *E. coli* isolates from 17 dogs and 6 cats. The identical isolates were

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classified as marbofloxacin-resistant in the MIC test [20].

However, ciprofloxacin should not be recommended for dogs because of low MIC values after oral administration. The MIC50 level observed in the presented study was 0.25 μg/mL. However, only 18% of the tested population of the dogs reached the effective concentration in serum (0,25 μg/mL) after oral administration of ciprofloxacin in a dose of 25 mg/kg, and 60% of the tested animals after a dose of 50 mg/kg once a day [35]. Administering ciprofloxacin 'off label' or 'extra label' in veterinary medicine should be avoided because of no legible registration.

An increase in resistance to aminopenicillins (ampicillin, amoxicillin with or without beta-lactamase inhibitors) and other beta-lactams among *E. coli* isolates due to its overuse or misuse in human and veterinary medicine appears to be alarming for public health [2, 36]. In the current study, 100% of *E. coli* isolates from feline urine were resistant to ampicillin and 54% to ampicillin with sulbactam. Among 66 isolates from dogs, 68% and 59% were resistant to ampicillin and ampicillin/sulbactam, respectively. Ampicillin resistance was documented in 35% of *E. coli* isolates in a study by Feßler et al. [20]. In the USA, no resistance to ampicillin was detected in 31.42% *E. coli* isolates, and the most common AMR resistance pattern was for ampicillin (18 isolates, 4.92%), chloramphenicol (16 isolates, 4.37%), and amoxicillin-clavulanic acid-ampicillin-cefazolin-cefovecin-cefpodoxime-ceftazidime-cephalexin (10 isolates, 2.73%) [8].

In the current study, the most frequent resistance patterns were (beta-lactams) ampicillin-ampicillin/sulbactam-ceftazidim-cefuroxime-aztreonam (14 isolates, 21.2% in dogs and 5 isolates, 12.2% in cats), ampicillin (4 isolates, 6,1% in dogs and 12 isolates, 29.3% in cats), beta-lactams and ciprofloxacin (4 isolates, 6.1% in dogs), ciprofloxacin (3 isolates, 4.5% in dogs), and ampicillin-ampicillin/sulbactam-ciprofloxacin (4 isolates, 9.8%) in cats. The results indicate a high percentage of strains resistant to beta-lactams. The high percentage of isolates' resistance to aztreonam (61% for dogs and 32% among cats) seems alarming. Martins et al. described the case of a dog with a history of urolithiasis and isolated a multidrug-resistant *E. coli*. The isolated bacteria were resistant to 13 antibiotics, being sensitive only to piperacillin-tazobactam and amikacin. The phenotypic resistance profile for ampicillin, amoxicillin-clavulanate, aztreonam, cefepime cefoxitin, cefuroxime, ceftazidime, ceftriaxone, imipenem, and piperacillin-tazobactam was identified by qPCR [37]. Among monobactams, aztreonam is one of the most resistant to metallo-β-lactamases (MBL) antimicrobials, and it is effective against gram-negative infections [38, 39]. Nordman et al., among 110 selected *E. coli* isolates with MIC >4 mg/mL to aztreonam have found 15 isolates resistant to aztreonam-avibactam, producing New Delhi metallo-beta-lactamase (NDM) and OXA-48-beta-lactamase. Isolates were collected between 2017 – 2019, however, the highest numbers were seen in 2019, suggesting recent emergence [40].

Resistance to aztreonam was documented for faecal *E. coli* isolates from 4 (6.4%) domestic dogs and 15 (30%) stray dogs in Argentina [41]. The similarity between the *E. coli* strains obtained from the urine and stool samples from this same patient seems to be significant. Bahadori et al., on the rate of similarity pulsed-field gel electrophoresis (PFGE) of urine and faecal *E. coli* isolates from the same patient, have documented genetically indistinguishable isolates (100%

similarity). Compared isolates were identical based on the virulence genes profile and antibiotic susceptibility pattern [42]. Therefore, urinary tract infections caused by aztreonam resistance isolates of *E. coli* in companion animals are very likely, as in the presented study. The lowest prevalence of resistance was documented for colistin (8% dogs, 10% cats) and gentamicin (14% dogs, 10% cats). Low resistance to colistin may be the result of the lack of registration for ready-to-use drugs containing colistin for dogs and cats. Another polymyxin, polymyxin B, because of its high toxicity, is used only topically in veterinary medicine [43]. Nonetheless, colistin is frequently considered a last-resort antibiotic due to its significance in the management of human diseases [44]. A systemic (intravenously, intramuscularly) administration of gentamycin carries a high risk of nephrotoxicity and ototoxicity. Even administering topically to the ear canal may lead to ototoxicity [45]. The mentioned risk causes a significant decrease in the usage of these drugs and may also lead to a reduction in resistance in tested *E. coli* isolates.

Age-related antimicrobial resistance was observed in some previous studies, both in veterinary and human medicine. Gaire et al. noticed mixed AMR dynamics with age among faecal microbiota in dogs and farm animals [46]. In studies carried out in Bangladesh among different age groups, patients with UTI presented higher resistance to amikacin, nitrofurantoin, and colistin. Resistance to meropenem increased with the age of the patients [47]. In the current study, a moderate positive correlation between age and multidrug-resistance was also observed.

Companion animals can be the host of multidrug resistant bacteria, which can be a potentially zoonotic infection. Naziri et al., among 28 dog and owner pairs, documented the same patterns of the presence of ESBL genes in 8 (28.6%) pairs and very similar resistance or susceptibility in the dog-owner pairs. Four dog-owner pairs had the same antimicrobial resistance patterns [48]. However, the zoonotic transmission was very likely. One of 10 UTI patients living with pets had the infection caused by this same strain, and in another case this same strain persisted in animals' faeces (not the owner) for 10 months [49].

CONCLUSIONS

Antibiotics belonging to the same chemical groups are used to treat infections both in animals and humans. The bacteria, including antimicrobial-resistant bacteria, may be transferred from animals to humans and from humans to animals by direct contact. Moreover, the antimicrobial-resistant bacteria may persist for up to several months without causing infection. Therefore, close contact with pets, increasing antimicrobial resistance, and very likely zoonotic potential of the same bacterial strains should be cause for alarm. Knowledge of antimicrobial resistance emergencies is important for public health, primary care physicians, veterinary clinicians, and pet owners. Preventive actions, such as cleaning pet bedding, washing animal bowls, and hand washing and sanitizing after petting animals, should be implemented to limit the spread of antimicrobial-resistant bacteria. Due to the to the limited number of antimicrobial resistant reports among bacterial strains isolated from dogs and cats urine in Poland, further research including other species recognized as uropathogens in companion

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animals are needed. The data from this study can be used to determine MIC breakpoints useful in clinical treatment of UTIs. Moreover, defined MIC breakpoints provide a basis for future MIC comparison.

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