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Research

Epidemiological Dynamics of Multidrug Resistance (Ampicillin, Streptomycin, and Tetracycline) in Domestic Cats in East Surabaya

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ABSTRACT

Antibiotic resistance in pets, especially cats, has become a serious global health challenge. The irrational use of antibiotics, whether excessive or inappropriate, accelerates the emergence of resistant bacteria that can potentially spread to humans and the environment. This study aims to identify the presence of antibiotic-resistant Escherichia coli and analyze its resistance patterns in domestic cats in East Surabaya. A total of 35 rectal swab samples were collected from pet cats and tested at the Veterinary Public Health Laboratory, Faculty of Veterinary Medicine, Wijaya Kusuma University, Surabaya. The samples were isolated using the selective MacConkey Agar medium and confirmed as Escherichia coli through Gram staining, biochemical tests, and antibiotic susceptibility testing. The results showed that 94.3% (33/35) of the samples contained Escherichia coli. Antibiotic susceptibility testing revealed resistance rates of 27% (9/33) to ampicillin, 24% (8/33) to streptomycin, and 12% (4/33) to tetracycline. Additionally, 9.1% (3/33) of the isolates were classified as multidrug resistance, showing resistance to all three antibiotics (AMP-TE-S). Other resistance patterns included AMP-S (6.1%) and TE-S (3.1%). The high resistance rate of Escherichia coli in domestic cats highlights the critical role of pets as potential reservoirs of resistant bacteria, which may impact human health through direct contact or environmental transmission. Therefore, raising awareness and educating pet owners and veterinary professionals on the prudent and responsible use of antibiotics is essential. The One Health approach, integrating human, animal, and environmental health, is a strategic measure to prevent the future spread of antimicrobial resistance.

Keywords: Escherichia coli, antibiotic resistance, multidrug resistance, domestic cats, Surabaya, One Health

ABSTRAK

Resistensi antibiotik pada hewan peliharaan, terutama kucing, telah menjadi tantangan serius dalam kesehatan global. Penggunaan antibiotik yang tidak rasional, baik secara berlebihan maupun tidak tepat sasaran, mempercepat munculnya bakteri resisten yang berpotensi menular ke manusia dan lingkungan. Penelitian ini bertujuan untuk mengidentifikasi keberadaan *Escherichia coli* yang resisten terhadap antibiotik serta menganalisis pola resistensinya pada kucing domestik di Surabaya Timur. Sebanyak 35 sampel swab anus dari kucing peliharaan dikumpulkan dan diuji di Laboratorium Kesehatan Masyarakat Veteriner, Fakultas Kedokteran Hewan, Universitas Wijaya Kusuma Surabaya. Sampel diisolasi menggunakan media selektif MacConkey Agar, kemudian dikonfirmasi sebagai *Escherichia coli* melalui pewarnaan Gram, uji biokimia, dan dilakukan uji sensitivitas untuk mengetahui adanya resistensi antibiotik. Hasil penelitian menunjukkan bahwa 94,3% (33/35) sampel mengandung *Escherichia coli* Uji resistensi antibiotik mengungkapkan bahwa 27% (9/33) isolat resisten terhadap ampisilin, 24% (8/33) terhadap streptomisin, dan 12% (4/33) terhadap tetrasiklin. Lebih lanjut, 9,1% (3/33) isolat dikategorikan sebagai *multidrug resistance* dengan pola resistensi terhadap ketiga antibiotik (AMP-TE-S), sementara pola resistensi lainnya meliputi AMP-S (6,1%) dan TE-S (3,1%). Tingginya tingkat resistensi *Escherichia coli* pada kucing domestik menyoroti peran penting hewan peliharaan sebagai reservoir potensial bakteri resisten, yang dapat berdampak pada kesehatan manusia melalui kontak langsung maupun lingkungan. Oleh karena itu, diperlukan kesadaran dan edukasi bagi pemilik hewan serta tenaga medis veteriner mengenai penggunaan antibiotik yang bijak dan bertanggung jawab. Pendekatan *One Health* yang mengintegrasikan kesehatan manusia, hewan, dan lingkungan menjadi langkah strategis dalam mencegah penyebaran resistensi antimikroba di masa depan.

Kata kunci: *Escherichia coli*, resistensi antibiotik, *multidrug resistance*, kucing domestik, Surabaya, *One Health*

INTRODUCTION

Antibiotic resistance is a growing global threat (CDC, 2019; Singh *et al*., 2019; Wibisono *et al*., 2024c), not only in humans (Salam *et al*., 2023) but also in livestock (Bourély *et al*., 2020; Nielsen *et al*., 2022), poultry (Wibisono *et al*., 2020a, 2020b), and domestic animals (Don Bamunusinghage *et al*., 2019; Li *et al*., 2021). Resistance in cats has been reported previously (Wibisono *et al*., 2024b, 2024d). Domestic cats (*Felis catus*) have the potential to become reservoirs of resistant bacteria that can impact animal and human health through zoonotic mechanisms (Das *et al*., 2023). Antibiotics such as streptomycin from the aminoglycoside group, ampicillin from the beta-lactam group, and tetracycline from the tetracycline group are antibiotics that are often used in the treatment of bacterial infections in animals, but uncontrolled use can encourage the emergence of multidrug resistance (MDR) patterns (Amelia *et al*., 2016; Rafiq *et al*., 2022). This increase in antibiotic resistance can hinder the effectiveness of treatment and worsen the health condition of animals. In addition, resistant bacteria found in beloved cats have the potential to spread to humans through direct contact, the environment, or food contamination, thereby posing a risk to public health (Das *et al*., 2023; Fanissa *et al*., 2022).

An epidemiological perspective shows that environmental factors, cat population density, and antibiotic usage patterns can influence the dynamics of antimicrobial resistance (Khairullah *et al*., 2023; Mandal *et al*., 2022; Samreen *et al*., 2021; Wibisono *et al*., 2024a). Urban areas such as East Surabaya, with high cat population density and close interaction with humans, provide an environment conducive to the selection and spread of resistant bacteria. Epidemiological studies on multidrug resistance patterns in domestic cats in this region are crucial for understanding the dynamics of resistance and its implications for public health. Therefore, this study aims to identify patterns of bacterial resistance to ampicillin, streptomycin, and tetracycline in domestic cats in East Surabaya, as a first step in mitigating and controlling antimicrobial resistance.

MATERIALS AND METHOD

Ethical approval

Ethical approval was obtained from the Animal Ethics Committee of the Faculty of Veterinary Medicine, Wijaya Kusuma University, Surabaya, with no. 123 - KKE. This study used 35 anal swab samples from domestic cats to isolate and identify resistant *Escherichia coli* bacteria. Domestic cats were obtained from owners in the eastern part of Surabaya. Samples were collected randomly without classification based on age, sex, or health status. The cats included were domestic cats, defined as pets living in close association with humans in household environments. This random sampling approach was applied to obtain a general overview of the presence of antibiotic-resistant *Escherichia coli* in the pet cat population.

Isolation and Identification of Escherichia coli

The samples were isolated and identified using MacConkey Agar (MCA), a differential selective medium containing special dyes and carbohydrates to distinguish colonies that ferment lactose (pink) from bacterial colonies that do not ferment lactose (colorless) (Akanbi *et al*., 2022). Bacteria were streaked in a zigzag pattern on the surface of MCA medium, then incubated at 37°C for 24-48 hours. Isolates identified as *Escherichia coli* were then subjected to Gram staining and biochemical testing (Wibisono *et al*., 2024c; Zikra *et al*., 2018).

The Triple Sugar Iron Agar (TSIA) test aims to differentiate Enterobacteriaceae bacteria. The TSIA test results on *Escherichia coli* isolates show a yellow color change on the slant and bottom media and the appearance of gas that causes the media to rise, but no H2S is visible (*Yuliandi et al*., 2022). The Simmon Citrate Agar (SCA) test is performed to differentiate enteric bacteria by observing their ability to use citrate as a carbon source. The citrate test results for *Escherichia coli* isolates were negative, as evidenced by the absence of citrate (Kendek *et al*., 2024). The urease test is used to distinguish bacteria based on their ability to hydrolyze urea using the urease enzyme. The results of the urease test on isolated strains of *Escherichia coli* are negative if the medium does not change color and remains orange, meaning that *Escherichia coli* cannot hydrolyze urea because it does not have the urease enzyme (Klaharn *et al*., 2022). The indole test is performed to determine the ability of bacteria to produce the amino acid tryptophan and will be indicated by the presence of indole. Positive indole test results for *Escherichia coli* strains are indicated by the presence of a pink ring after the addition of Kovac's reagent. The motility test is performed to determine bacterial motility. Positive results are indicated by the formation of a cloud in the puncture area. (Rahayu & Gumilar, 2017). The Methyl Red (MR) test is performed to determine the ability of bacteria to

ferment mixed acids. The MR test results on *Escherichia coli* isolates showed positive results due to a red color change in the medium. The Voges Proskaer (VP) test was conducted to determine the ability of a bacterium to produce acetylmethylcarbinol or acetoin. The VP test results on *Escherichia coli* isolates showed negative results, indicated by no color change in the medium, meaning that *Escherichia coli* cannot produce acetoin (Wibisono *et al.*, 2024d)

Sensitivity Test

Antibiotic sensitivity testing is a test conducted to determine the sensitivity of bacteria to antibiotics. The sensitivity of a bacterium to antibiotics is determined by the diameter of the inhibition zone formed. The larger the diameter of the inhibition zone, the more inhibited the growth, so a reference standard is needed to determine whether the bacterium is resistant or sensitive to an antibiotic (CLSI, 2022).

Data Analysis

The data analysis method in this study included antibiotic sensitivity testing using the disk diffusion method (Kirby-Bauer) to assess bacterial resistance patterns to various classes of antibiotics. The data obtained were analyzed descriptively to determine the incidence of multidrug resistance (MDR) in domestic cats in the East Surabaya area, and analyzed using the Chi-square test to determine differences in the proportion of *Escherichia coli* isolates that were sensitive or resistant among sampling areas, and to analyze the relationship between the ownership-based housing patterns of cats, specifically cats allowed to roam outdoors and those kept strictly indoors, with a significance level of 5% ($\alpha = 0.05$).

RESULTS

Isolation and Identification of Escherichia coli

The results of isolation and identification of a total of 35 samples collected from cat anal swabs showed a result of 94.3% (33/35), colonies with red, round, and dry morphology. Macroscopic examination revealed distinct characteristics, including circular colonies with smooth edges, convex surfaces, and a reddish-brown color (Figure 1, point A). Gram staining to determine microscopic morphology using 1000x magnification showed results with short rod-shaped morphology (Coccobacillus) and red coloration. These results can be classified as Gram-negative bacteria, with the red color observed being safranin, the secondary stain in Gram staining (Figure 1, point B).

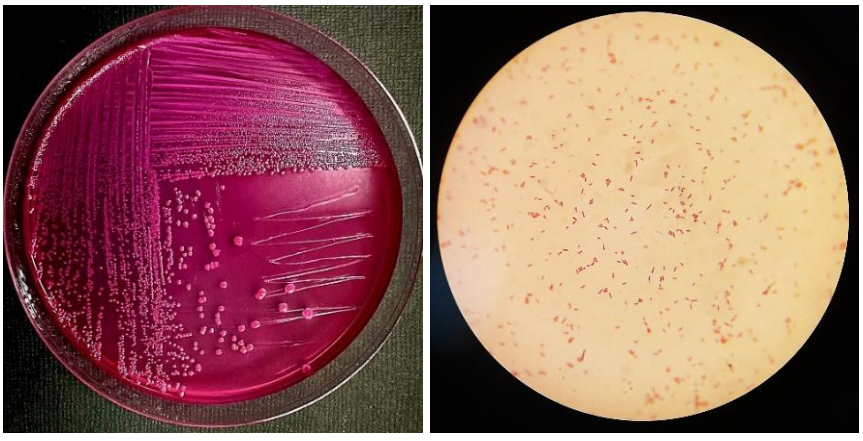

Figure 1. Colony of *Escherichia coli* bacteria on MacConkey Agar (MCA) medium (A), Microscopic morphology of *Escherichia coli* bacteria stained with Gram stain (B).

Sensitivity Test

The results of sensitivity testing for *Escherichia coli* in stray cats in East Surabaya were conducted on Muller Hilton Agar (MHA) media, showing clear zones or inhibition zones around the antibiotic discs (Figure 2). The antibiotics used were ampicillin, tetracycline, and streptomycin. The results of the study showed that *Escherichia coli* isolated from domestic cats in the East Surabaya area had varying levels of resistance to several types of antibiotics tested. Ampicillin had the highest resistance rate of 27% (9/33), followed by streptomycin at 24% (8/33), and tetracycline at 12% (4/33) (Figure 3, Table 1).

Multidrug Resistance

The results of sensitivity testing show that if there are three or more types of antibiotics from different groups that show resistance, then the antibiotic is categorized as multidrug resistant. The results of the study show that 9.1% (3/33) of *Escherichia coli* isolates from domestic cats in East Surabaya are classified as MDR (Table 2), which are resistant to the three types of antibiotics tested: ampicillin, streptomycin, and tetracycline (Figure 2). The presence of these MDR isolates indicates significant selection pressure on *Escherichia coli*, possibly due to the uncontrolled use of antibiotics in animal treatment. *Chi-square Analysis of Antibiotic Resistance among Sampling Areas.*

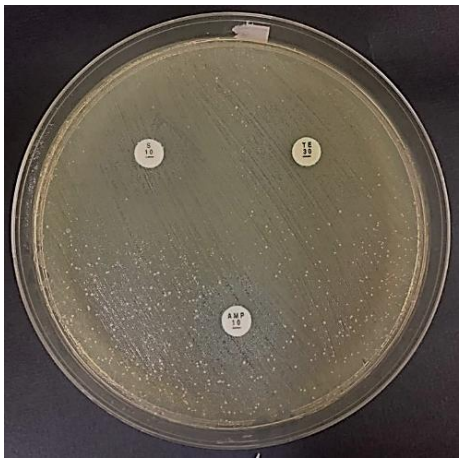

Figure 2. Sensitivity test of *Escherichia coli* bacteria (TE = Tetracycline, AMP = Ampicillin, S = Streptomycin). Multidrug resistant (A), not multidrug resistant (B)

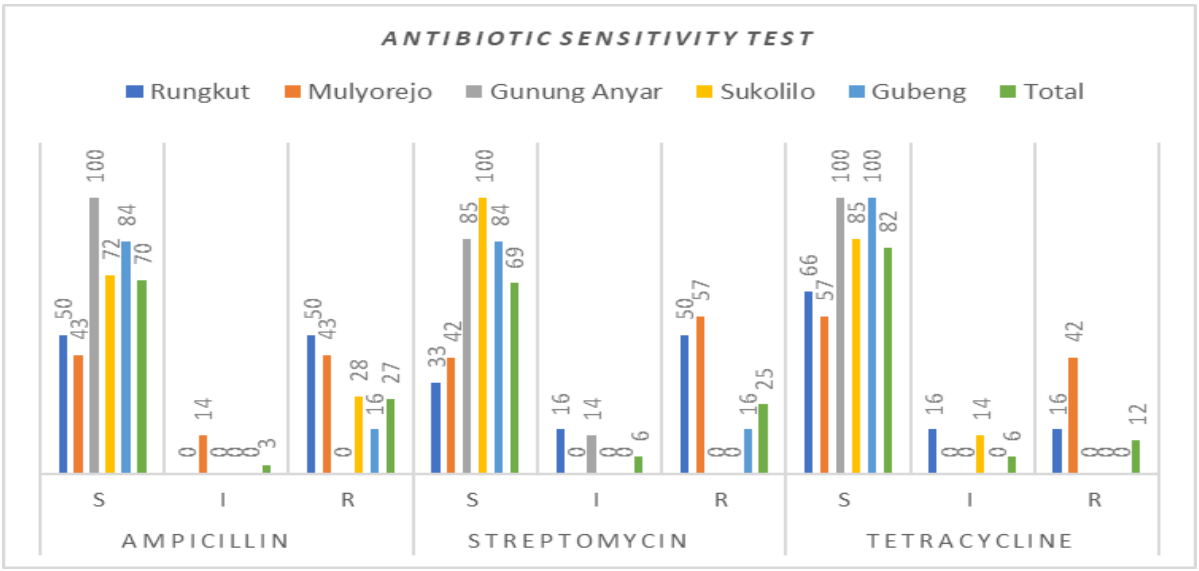

Figure 3. Antibiotic Sensitivity Test in Escherichia coli

Table 1 Antibiotic Sensitivity Test

	Ampicillin			Streptomycin			Tetracycline		
	S	**I**	**R**	**S**	**I**	**R**	**S**	**I**	**R**
				East Surabaya					
Rungkut	50% (3/6)	0% (0/6)	50% (3/6)	33% (2/6)	16% (1/6)	50% (3/6)	66% (4/6)	16% (1/6)	16% (1/6)
Mulyorejo	42% (3/7)	14% (1/7)	42% (3/7)	42% (3/7)	0% (0/7)	57% (4/7)	57% (4/7)	0% (0/7)	42% (3/7)
Gunung Anyar	100% (7/7)	0% (0/7)	0% (0/7)	85% (6/7)	14% (1/7)	0% (0/7)	100% (7/7)	0% (0/7)	0% (0/7)
Sukolilo	71% (5/7)	0% (0/7)	28% (2/7)	100% (7/7)	0% (0/7)	0% (0/7)	85% (6/7)	14% (1/7)	0% (0/7)
Gubeng	83% (5/6)	0% (0/6)	16% (1/6)	83% (5/6)	0% (0/6)	16% (1/6)	100% (6/6)	0% (0/6)	0% (0/6)
Total	**69 % (23/33)**	**3 % (1/33)**	**27 % (9/33)**	**69 % (23/33)**	**6 % (2/33)**	**24 % (8/33)**	**81 % (27/33)**	**8 % (2/33)**	**12 % (4/33)**

Table 2 Patterns of Resistance in *Escherichia coli* Bacteria

No	Resistance Patterns	percentage
1	AMP-TE-S	9.1% (3/33)
2	AMP-TE	0% (0/33)
3	AMP-S	6.1% (2/33)
4	TE-S	3.1% (1/33)
5	AMP	12.1% (4/33)
6	TE	0% (0/33)
7	S	3.1% (1/33)

The Chi-square test was conducted to evaluate whether there were differences in the proportion of *Escherichia coli* isolates that were sensitive or resistant among the five sampling areas (Rungkut, Mulyorejo, Gunung Anyar, Sukolilo, and Gubeng). The analysis yielded a Chi-square value (χ^2) of 7.0714 (ampicillin), 7.0662 (streptomycin), and 6.7513 (tetracycline), with 4 degrees of freedom (χ^2 tabel 9.488) and a p-value of 0.1132 (ampicillin), 0.1234 (streptomycin), and 0.1496 (tetracycline). Since the p-value was greater than the significance threshold ($p > 0.05$), no statistically significant difference was observed in the distribution of resistant and sensitive isolates among the sampling areas.

This indicates that the prevalence of antibiotic-resistant *Escherichia coli* was relatively similar across all study locations (Table 3).

Chi-square Test Results on Cat Keeping Patterns

The Chi-square analysis showed that the pattern of cat keeping (outdoor vs. indoor) was significantly associated with antibiotic resistance in *Escherichia coli* for streptomycin ($\chi^2 = 10.7$; $p = 0.00$). Cats allowed outdoors were found to have 16 times higher odds (OR = 16) and three times higher risk (RR = 3) of carrying streptomycin-resistant *Escherichia coli* compared to indoor cats. In contrast, no significant associations were observed for tetracycline ($\chi^2 = 2.73$; $p = 0.10$) and ampicillin ($\chi^2 = 1.47$; $p = 0.23$), indicating that resistance to these antibiotics was not influenced by the cat keeping pattern (Table 4). These results suggest that cats with outdoor access are more likely to be exposed to environmental bacterial sources or other animals carrying resistant E. coli, thereby increasing the risk of colonization by resistant strains.

Conversely, indoor cats, which have limited environmental exposure, tend to have a lower likelihood of harboring antibiotic-resistant bacteria. This finding highlights the role of environmental exposure and animal movement in the spread of antimicrobial resistance among companion animals.

DISCUSSION

Isolation and Identification of Escherichia coli

The isolation and identification results showed that *Escherichia coli* was successfully isolated from domestic cat samples in East Surabaya, confirming that this bacterium is common microflora in the digestive tract of cats. *Escherichia coli* is known as a commensal bacterium that can become an opportunistic pathogen, especially when there is an imbalance in the microbiota or selective pressure due to the use of antibiotics (Gambino *et al*., 2023).

The identification process was carried out through biochemical tests, including indole, methyl red, Voges-Proskauer, and citrate (IMVIC) tests, which showed the characteristic features of *Escherichia coli*, such as the ability to produce indole and ferment glucose with acid production without gas production. Additionally, colony selection results on differential media such as MacConkey agar confirm the presence of *Escherichia coli* through the growth of pink colonies, which indicates lactose fermentation (Imasari & Ula, 2023).

Sensitivity Test

Ampicillin is the antibiotic of choice as first-line therapy for treating *Escherichia coli* infections in cats and dogs. This results in a higher percentage of ampicillin resistance compared to tetracycline and streptomycin (Awosile *et al.,* 2018). The high level of resistance to ampicillin is consistent with previous

Table 3 Chi-square Analysis of Antibiotic Resistance among Sampling Areas

No	Antibiotic	Chi-square (χ^2)	df	p-value	Result
1	Ampicillin	7.0714	4	0.1132	no statistically significant (p > 0.05)
2	Streptomycin	7.0662	4	0.1234	no statistically significant (p > 0.05)
3	Tetracycline	6.7513	4	0.1496	no statistically significant (p > 0.05)

Table 4 *Chi-square Test Results on Cat Keeping Patterns*

No	Antibiotic	Variable	D+	D-	Chi-square (χ^2)	df	p-value	Result	OR/RR
1	Ampicillin	Outdoor Indoor	18 5	5 5	1.47	2	0.23	no statistically significant (p > 0.05)	-
2	Streptomycin	Outdoor Indoor	20 3	3 7	8.18	2	0.00*	statistically significant (p < 0.05)	OR 16 RR 3
3	Tetracycline	Outdoor Indoor	21 6	2 4	2.73	2	0.1	no statistically significant (p > 0.05)	-

studies showing that *Escherichia coli* often develops resistance to beta-lactam antibiotics due to the production of beta-lactamase enzymes, including Extended-Spectrum Beta-Lactamases (ESBL). Another factor that may contribute to this high level of resistance is the widespread use of antibiotics, both in the treatment of domestic animals and in the surrounding environment, which allows for the selection of resistant bacteria (Gargano *et al*., 2022; Wibisono *et al*., 2024c).

Streptomycin resistance reaching 24% indicates the possible accumulation of resistance genes, such as the *aad*A gene, which is often found in plasmids or transposons and can be transferred between bacteria through horizontal gene transfer mechanisms. The use of streptomycin in veterinary practice as the antibiotic of choice for gastrointestinal and respiratory tract infections may also contribute to this increase in resistance (Castanheira *et al*., 2021; Normaliska *et al*., 2019; Shafiq *et al*., 2021).

Meanwhile, lower resistance to tetracycline (12%) indicates that although this antibiotic is still effective against most *Escherichia coli* isolates, monitoring of its use is still necessary. Resistance to tetracycline is generally associated with the presence of the tet(A) or tet(B) genes, which encode efflux pump mechanisms that reduce the concentration of antibiotics within bacterial cells (García-béjar *et al*., 2021; Widhi & Saputra, 2021).

Multidrug Resistance

Based on resistance patterns, the percentage of isolates exhibiting multidrug resistance was 9.1% (3/33), code STP24 (Gunung anyar), KRR7 (Gubeng), KRR11 (Gubeng), namely to the antibiotic ampicillin, tetracycline, and streptomycin. MDR in *Escherichia coli* is often caused by multiple defense mechanisms, such as the production of beta-lactamase enzymes to counter ampicillin, modification of the ribosomal target against streptomycin, and efflux pump systems for tetracycline. The combination of these mechanisms increases bacterial resistance to various classes of antibiotics, thereby limiting effective treatment options (Chen *et al*., 2019; Rzewuska *et al*., 2015).

Previous studies have shown that feeding raw food to cats is associated with a higher risk of infection with MDR and ESBL-producing *Escherichia coli* compared to cats fed dry food. This remarkable adaptability of commensal *Escherichia coli* to the environment as a result of antibiotic intervention can mainly be attributed to an effective elimination system.

This system is the first line of defense for *Escherichia coli* and can therefore be considered part of the innate resistance mechanism against antibiotics. As a result, these bacteria adapt to new antibiotics (e.g., streptomycin or tetracycline) within hours of treatment and respond with an increase in population that may become dominant in the normal intestinal flora for several days and, depending on the antimicrobial used, may remain at a minority level of the normal *Escherichia coli* state later on (Fayez *et al*., 2023).

The high prevalence of MDR, although still relatively small in percentage terms, is a serious concern because it can contribute to the spread of antimicrobial resistance in the environment, including the potential for transmission to humans through direct contact with domestic animals. This is in line with the One Health concept, which emphasizes the interconnection between human, animal, and environmental health in addressing antimicrobial resistance.

Chi-square Analysis of Antibiotic Resistance among Sampling Areas

The Chi-square test results showed no statistically significant differences in the proportion of antibiotic-resistant *Escherichia coli* isolates among the five sampling areas (Rungkut, Mulyorejo, Gunung Anyar, Sukolilo, and Gubeng) for ampicillin, streptomycin, and tetracycline ($p > 0.05$). This finding indicates that the prevalence of antibiotic resistance in *E. coli* isolated from domestic cats was relatively similar across the study locations.

The absence of significant variation among areas suggests that environmental exposure, antibiotic usage practices, and cat-keeping behaviors may be relatively homogeneous across these regions. Domestic cats in urban environments such as Surabaya often share similar ecological and social conditions, including close contact with humans, exposure to similar food sources, and access to veterinary care. These factors could contribute to the uniform pattern of antibiotic resistance observed.

Although no regional difference was detected, the presence of resistant *E. coli* isolates in all areas highlights the widespread nature of antimicrobial resistance (AMR) within companion animal populations. This finding aligns with previous studies reporting that household pets can serve as reservoirs and potential disseminators of antibiotic-resistant bacteria within urban communities.

Chi-square Test Results on Cat Keeping Patterns

The results of this study demonstrated that the pattern of cat keeping was significantly associated with *Escherichia coli* resistance to streptomycin, but not to ampicillin or tetracycline. Cats that were allowed to roam outdoors showed a 16 fold higher odds and a threefold higher relative risk of carrying streptomycin-resistant *Escherichia coli* compared to cats kept strictly indoors. This finding indicates that environmental exposure plays a crucial role in the acquisition and dissemination of antimicrobial-resistant bacteria among domestic cats.

Outdoor cats are more likely to encounter various environmental sources of resistant bacteria, such as contaminated soil, water, or contact with other animals, including stray cats or wildlife. These interactions increase the opportunity for horizontal transfer of resistance genes, especially through mobile genetic elements such as plasmids.

Streptomycin resistance is often associated with aminoglycoside-modifying enzymes encoded on plasmids, which can be easily exchanged among bacteria in diverse ecological niches. In contrast, no significant association was observed for ampicillin and tetracycline resistance, suggesting that these resistance determinants may already be widespread in both outdoor and indoor environments. Ampicillin and tetracycline are among the most commonly used antibiotics in veterinary and human medicine, which may have led to a baseline prevalence of resistant *E. coli* across different cat populations regardless of their housing patterns.

Reinforced by previous research, the possibility of antibiotic resistance in *Escherichia coli* allows it to be transmitted from domestic animals such as dogs and cats to humans, either directly or indirectly, because they share the same environment as humans, are in close proximity, and are also exposed to antibiotics for therapeutic purposes, which are commonly used for humans. Antibiotic resistance is based on the nature of bacteria and how antibiotics work according to their class and target in killing bacteria (Moon *et al., 2023*).

The successful isolation of *Escherichia coli* from domestic cats shows that these animals can serve as potential reservoirs for bacteria, including strains that have the potential for antibiotic resistance. Environmental conditions, dietary patterns, and interactions with humans and other animals can influence the composition and level of bacterial resistance present in cats. Therefore, monitoring the presence and characteristics of *Escherichia coli* resistance is important, especially in the context of animal health and zoonotic risks.

The present study revealed a high level of *Escherichia coli* resistance to ampicillin in domestic cats from the Surabaya area (Wibisono *et al.*, 2024d). This phenomenon can be explained by several local factor characteristic of the urban environment in Surabaya. Firstly, the high density of domestic cat populations that frequently interact within densely populated neighborhoods increases the likelihood of resistant bacteria transmission, either through direct contact or environmental contamination. Secondly, the uncontrolled use of antibiotics by pet owners, such as the administration of amoxicillin or ampicillin without veterinary prescription, likely contributes to the natural selection of resistant *Escherichia coli* strains. In addition, the humid and less hygienic conditions common in urban settings, particularly around waste disposal sites and drainage channels, may also serve as reservoirs for resistant bacteria.

To reduce the risk of spreading MDR bacteria, preventive measures are needed, such as the prudent use of antibiotics, the implementation of good hygiene in animal husbandry, and stricter supervision of the distribution and use of antibiotics in the veterinary sector. In addition, regular monitoring of resistance patterns is essential to identify resistance trends and adjust control strategies to be more effective in the future. The results of this study underscore the importance of rational antibiotic use and strict monitoring of antimicrobial resistance, especially in domestic animals that can be reservoirs for resistant bacteria that have the potential to be transmitted to humans (zoonoses). Mitigation measures are needed, such as increasing animal owners' awareness of the prudent use of antibiotics and implementing antimicrobial resistance control policies in animal health.

From the results of this study, it can be concluded that *Escherichia coli* isolated from domestic cats in East Surabaya showed an alarming pattern of resistance to several commonly used antibiotics. Resistance testing results show that ampicillin has the highest resistance rate (27%), followed by streptomycin (24%) and tetracycline (12%), reflecting the selective pressure caused by uncontrolled antibiotic use. In addition, 9.1% of *Escherichia coli* isolates were identified as multidrug

resistant, meaning that the bacteria were resistant to the three types of antibiotics tested. This indicates the potential threat of antimicrobial resistance that could impact animal and human health, especially given the role of cats as reservoirs of bacteria that can spread resistance through direct contact and the environment.

The findings of this study have important practical implications for antimicrobial resistance(AMR) control in Surabaya. The high levels of *Escherichia coli* resistance to ampicillin, streptomycin, and tetracycline in domestic cats highlight the urgent need for stricter antibiotic stewardship policies at the municipal level. Local authorities, together with veterinary health agencies, can use these results as a foundation for reinforcing regulations on antibiotic use in companion animals, including the implementation of mandatory prescription systems and restrictions on over-the-counter veterinary antibiotic sales.

Furthermore, education for cat owners plays a crucial role in mitigating resistance. Owners should be informed about the risks of administering antibiotics without veterinary supervision, the importance of completing prescribed treatments, and the need to maintain proper hygiene in areas where cats live and interact to prevent the spread of resistant bacteria.

In addition, these findings emphasize the necessity of adopting a *One Health* approach by integrating resistance surveillance across animals, humans, and the environment. Establishing a coordinated surveillance system in Surabaya, linking veterinary laboratories, public health facilities, and environmental agencies, would provide a more comprehensive understanding of AMR dissemination patterns. Such cross-sectoral collaboration not only supports the control of resistance in companion animals but also safeguards public health and preserves the long-term efficacy of antibiotics.

"The author(s) report no conflicts of interest in this work".

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