

## Pathogenicity and antibiotic sensitivity of *Escherichia coli* isolates from non-diarrhoeic dogs

Nadira Seepersadsingh, and Abiodun A. Adesiyun\*

School of Veterinary Medicine, Faculty of Medical Sciences, University of the West Indies, St. Augustine, Trinidad and Tobago

---

**SEEPERSADSINGH, N., A. A. ADESIYUN: Pathogenicity and antibiotic sensitivity of *Escherichia coli* isolates from non-diarrhoeic dogs. Vet. arhiv 77, 531-542, 2007.**

### ABSTRACT

*E. coli* isolated from dogs in Trinidad were tested for their susceptibility to antimicrobial agents using the disk diffusion method. Antimicrobial agents and concentrations included cephalothin (KF, 30 µg), ampicillin (AMP, 10µg), kanamycin (K, 30 µg), neomycin (N, 30 µg), gentamicin (CN, 10 µg), sulphamethoxazole/trimethoprim (SXT, 23.25 µg/1.75 µg), nalidixic acid (NA, 30 µg) and norfloxacin (NOR, 10 µg). The overall prevalence of resistance to one or more antimicrobial agents for *E. coli* isolated from dogs was 47.9%. The difference in prevalence across the various sources of the isolates from dogs was statistically significant ( $P < 0.001$ ;  $\chi^2$ ). Overall, resistance was highest to cephalothin (30.1%). A total of 45 resistance patterns were observed from dogs from all sources and the predominant pattern was KF (25.6%). It was concluded that the relatively high prevalence of resistance to antimicrobial agents amongst *E. coli* isolates from non-diarrhoeic dogs in Trinidad may pose zoonotic and therapeutic problems.

**Key words:** antimicrobial sensitivity, *Escherichia coli*, non-diarrhoeic dogs

---

### Introduction

Antibiotics are used to treat microbial infections in humans and animals as well as being given prophylactically to prevent infections (ESPINASSE, 1993). They are also given in low doses to food animals to improve their growth rate (HELMUTH, 2000). When bacterial populations are exposed to antimicrobial substances, the possibility exists that there may be an emergence of resistance (STERNBUERG, 1999). Bacteria acquire resistance to antimicrobial drugs in response to a wide range of selection pressures which may operate in different ways but not always clearly identifiable (NORMAND et al., 2000).

---

\*Contact address:

Professor A. A. Adesiyun, School of Veterinary Medicine, Faculty of Medical Sciences, University of the West Indies, St. Augustine, Trinidad and Tobago, Phone: +1868 645 8329; +1868 645 4481; +1868 777 7480; Fax.: +1868 645 7428; E-mail: aadesiyun@fms.uwi.tt; abiodunadesiyun@hotmail.com

The use of antibiotics itself leads to a selection for antimicrobial resistant isolates, and this resistance can be disseminated by the spread of the bacteria or by transfer of genes to other bacteria (ANONYMOUS, 1998). It is well recognized today that resistance genes can be exchanged among bacteria populations (DAVIES, 1998).

Pets are, by definition, in close contact with people, and there is ample opportunity for exchange of resistance genes between bacteria from these different host species. Dogs are probably pets to which most antimicrobial agents are administered. The antimicrobial substances used in dogs are often similar, or identical, to those used in human medicine (STERNBUERG, 1999). Bacteria with potentially transferable antimicrobial resistance determinants have been reported to be isolated from rectal swabs taken from healthy dogs in urban and rural environments (ANONYMOUS, 1998; NORMAND et al., 2000). DAVIES et al. (1978) reported the interrelationship of antimicrobial resistance in the flora of humans and domestic pets and concluded the resistance plasmids in the two populations to be similar.

Heavy use of antibiotics in settings such as hospitals and farms, where drugs are often given to animals to enhance growth, may increase the level of resistant bacteria (LEVY, 1998). Hospitalized animals are frequently exposed to an environment laden with antimicrobial substances which may facilitate the transmission of resistance genes (STERNBUERG, 1999). Resistant hospital-acquired organisms can also be spread to community contacts as in walk-in clinics or companion-animal community practices that also serve as hospitals (DECKER and SCHAFFNER, 1992).

The objective of this study was to determine the antimicrobial susceptibility patterns of *E. coli* isolates from dogs from various sources.

## **Materials and methods**

*Types and sources of samples.* The types of samples collected were either rectal or fresh faeces. All samples were taken aseptically. All samples taken originated from non-dairrhoaic animals. The source of samples included dogs from households; the Dog pound which represented the stray population; The Trinidad and Tobago Society for the Prevention of Cruelty to Animals (TTSPCA) served as the source of samples from an animal shelter; Veterinary establishments served as a source of samples from both inpatients and outpatients; The Quarantine Station served as the source of dogs from foreign countries; Two major dairy farming areas (Waller Field and Carlsen Field served as sources of dogs living on dairy farms with exposure to dairy animals); Hunting dogs represented animals exposed to wildlife in their activities.

*Sample collection and transportation.* Samples taken as rectal swabs employed the use of sterile cotton-tipped applicators. Swabs were inserted into the rectum of the animal,

rotated gently and then placed in tubes containing sterile Amies Transport Medium (ATM) (Difco, Detroit, U.S.A). Samples taken in the form of fresh faeces were put in sterile plastic faecal cups using sterile wooden tongue depressors. All samples were transported ice-cooled to the laboratory within 24 h of collection.

*Questionnaires.* Questionnaires were specifically prepared for each source of samples studied.

*Isolation and identification of Escherichia coli.* For the initial isolation of *E. coli*, eosin methylene blue (EMB) agar (Oxoid, Basingstoke, U.K.) plates were used. Rectal swabs or fresh faeces were streaked for isolation on the EMB agar plates. Plates were then incubated overnight at 37 °C. Morphologically different colonies exhibiting a metallic green sheen were then picked for biochemical identification using standard methods (MACFADDIN, 1980). Identified isolates of *E. coli* were inoculated onto blood agar plates (BAP) (Oxoid, Basingstoke, U.K.) and incubated at 37 °C overnight to determine ability to produce hemolysins and mucoid colonies. Colonies from BAP were also subcultured onto Sorbitol MacConkey (SMAC) (Oxoid, Basingstoke, U.K.) and incubated at 37 °C overnight. Pale or colourless colonies were classified as non-sorbitol fermenters (NSF) while pink colonies were classified as sorbitol fermenters (SF) as earlier recommended (MARCH and RATNAM, 1986).

*Detection of O157 isolate of E. coli.* Isolates of *E. coli* that were non-sorbitol fermenting on SMAC were subjected to a slide and tube agglutination test using commercially prepared O157 *E. coli* antiserum (Difco, Detroit, U.S.A) (ØRSHOV and ØRSHOV, 1984).

*Detection of enteropathogenic isolates of E. coli.* The slide agglutination test was used to determine *E. coli* isolates which belonged to enteropathogenic serogroups using *E. coli* polyvalent antisera A, B and C (S. A. Scientific., Texas, U.S.A.). A total of 411 isolates of *E. coli* were tested from the various sources. The number of isolates selected to determine the presence of enteropathogenic serogroups was proportional to the total number of *E. coli* isolates recovered from each source. Random selection was thereafter used to determine the allotted number of isolates to be tested within each source.

*Detection of verocytotoxigenic (VT) isolates of E. coli.* A cell culture technique employing Vero cells from the kidney of the African Green Monkey was used to determine the verocytotoxigenicity of *E. coli* isolates (KONOWALCHUK et al., 1977).

*Antibiotic resistance of E. coli.* The Kirby Bauer disk diffusion method (BAUER et al., 1966) was used to determine antimicrobial resistance of the *E. coli* isolates tested. All isolates from dogs which exhibited virulence markers, specifically, hemolysin production (81 isolates), mucoid colonies (100), non-sorbitol fermenters (133), enteropathogenicity (187) and verocytotoxigenicity (74) singly or in combination, were subjected to antimicrobial sensitivity testing. *E. coli* isolates negative for virulence markers (125

isolates) were also tested for their sensitivity to these antimicrobial agents. The number of virulence marker-negative isolates was proportional to the total number of *E. coli* isolates recovered from each source. Random selection was thereafter used to determine the allotted number of isolates to be tested within each source.

Antimicrobial agents and concentrations used in the study were determined by information provided by local veterinarians as to the commonly used agents in the country. These included cephalothin (KF, 30 µg), ampicillin (AMP, 10 µg), kanamycin (K, 30 µg), neomycin (N, 30 µg), gentamicin (CN, 10 µg), nalidixic acid (NA, 30 µg), norfloxacin (NOR, 10 µg) and sulphamethoxazole/trimethoprim (SXT, 23.25 µg/1.75 µg). The susceptibility of the *E. coli* isolates to the various antimicrobial agents was read and compared to the zone sizes stipulated on the table provided by the disk manufacturer.

*Statistical analysis of data.* The chi-square test for independence was employed to compare prevalences and the type 1 error was set at 0.05 for all tests. Epi-Info (Center for Disease Control and Prevention, Atlanta, Georgia, U.S.A; Version 6.02) was used to determine whether there were any statistically significant differences in the parameters investigated.

## Results

The prevalence of resistance to selected antimicrobial agents amongst *E. coli* isolates from various sources is shown in Table 1. Overall, of the 645 *E. coli* isolates from dogs tested for antimicrobial resistance, 309 (47.9%) were resistant to one or more of the eight antimicrobial agents tested. The difference in prevalence across the various sources of the isolates was statistically significant ( $P < 0.001$ ;  $\chi^2$ ). Prevalence ranged from 23.2% in dairy farm dogs to 74.7% in hospitalized dogs. Amongst all isolates tested, resistance was highest to cephalothin (30.1%) and lowest to norfloxacin (0.5%).

For the eight antimicrobial agents tested, *E. coli* isolates from hospitalized dogs displayed the highest prevalence of resistance to 4 antimicrobial agents (KF, AMP, SXT and NA) compared with all the other sources. Six (35.3%) of the 17 isolates of *E. coli* that exhibited resistance to gentamicin were recovered from hospitalized dogs. Of the three *E. coli* isolates resistant to norfloxacin, one (33.3%) was recovered from a hospitalized dog, one (33.3%) from a clinic dog, and the third (33.3%) from a quarantine dog.

Regarding closeness to owners amongst dogs from households (dairy farms, hunting and households), of the 103, 110 and 12 *E. coli* isolates from dogs that experienced close, moderate and no contact respectively, 39 (37.9%), 47 (42.7%) and 5 (41.7%) were resistant to one or more of the eight antimicrobial agents tested (Table 2). The difference in resistance according to human contact was not statistically significant ( $P > 0.05$ ;  $\chi^2$ ). Similarly, for dogs kept under kennel conditions (Table 2), no significant difference

Table 1. Prevalence of resistance to antimicrobial agents amongst *E. coli* isolates from dogs

Source of samples	N° of animals sampled	N° of <i>E. coli</i> isolates	N° of <i>E. coli</i> isolates tested	N° (%) of isolates resistant <sup>a</sup>	N° (%) of <i>E. coli</i> isolates resistant to:							
					KF <sup>b</sup>	AMP	SXT	K	N	NA	CN	NOR
Quarantine	125	311	112	44 (39.2)	20 (17.9)	28 (25.0)	11 (9.8)	6 (5.4)	2 (1.8)	2 (1.8)	0 (0.0)	1 (0.9)
Households	350	429	122	56 (45.9)	43 (35.2)	10 (8.2)	4 (3.3)	19 (15.6)	5 (4.1)	4 (3.3)	3 (2.5)	0 (0.0)
Hospitals <sup>c</sup>	181	239	99	74 (74.7)	39 (39.4)	58 (58.6)	34 (34.3)	16 (16.2)	9 (9.1)	8 (8.1)	6 (6.1)	1 (1.0)
Pound	252	282	83	39 (46.9)	29 (34.9)	10 (12.1)	4 (4.8)	16 (19.3)	10 (12.1)	2 (2.4)	0 (0.0)	0 (0.0)
TTSPCA <sup>d</sup>	119	176	70	34 (48.6)	25 (35.7)	10 (14.3)	7 (10.0)	12 (17.1)	7 (10.0)	2 (2.9)	5 (7.1)	0 (0.0)
Dairy farms <sup>e</sup>	110	148	56	13 (23.2)	12 (21.4)	2 (3.6)	1 (1.8)	1 (1.8)	0 (0.0)	1 (1.8)	1 (1.8)	0 (0.0)
Clinics <sup>f</sup>	146	175	53	27 (50.9)	15 (28.3)	14 (26.4)	10 (18.9)	9 (16.9)	9 (16.9)	1 (1.9)	2 (3.8)	1 (1.9)
Hunting dogs	100	134	47	22 (46.8)	11 (23.4)	12 (25.5)	5 (10.6)	8 (17.0)	5 (10.6)	3 (6.4)	0 (0.0)	0 (0.0)
Pet shops	8	6	3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Total	1391	1900	645	309 (47.9)	194 (30.1)	144 (22.3)	76 (11.8)	87 (13.5)	47 (7.3)	23 (3.6)	17 (2.6)	3 (0.5)

<sup>a</sup>Resistant to one or more antimicrobial agents, <sup>b</sup>KF - Cephalothin, AMP - Ampicillin, SXT - Sulphamethoxazole/Trimethoprim, K - Kanamycin, N - Neomycin, NA - Nalidixic acid, CN - Gentamicin, Nor-Norfloraxacin, <sup>c</sup>In-patients, <sup>d</sup>Trinidad and Tobago Society for the Prevention of Cruelty to Animals; <sup>e</sup>Include Waller Field (25) and Carlsen field (25); <sup>f</sup>Include Waller Field (25) and Carlsen field (25); <sup>g</sup>Out-patients

Table 2. Prevalence of resistance to antimicrobial agents amongst *E. coli* isolates from dogs according to closeness to owners, kenneled sources and veterinary establishments

	N° of <i>E. coli</i> isolates tested	N° (%) of isolates resistant <sup>a</sup>	N° (%) of <i>E. coli</i> isolates resistant to:							
			KF <sup>b</sup>	AMP	SXT	K	N	NA	CN	NOR
Human contact with household <sup>c</sup> dogs										
Close	103	39 (37.9)	30 (29.1)	8 (7.8)	4 (3.9)	14 (13.6)	3 (2.9)	3 (2.9)	2 (1.9)	0 (0.0)
Moderate	110	47 (42.7)	34 (30.9)	15 (13.6)	6 (5.5)	13 (11.8)	7 (6.4)	3 (2.7)	2 (1.8)	0 (0.0)
Non-existent	12	5 (41.7)	2 (16.7)	1 (8.3)	0 (0.0)	1 (8.3)	0 (0.0)	2 (16.7)	0 (0.0)	0 (0.0)
Total	225	91 (40.4)	66 (29.3)	24 (10.7)	10 (4.4)	28 (12.4)	10 (4.4)	8 (3.6)	4 (1.8)	0 (0.0)
Kenneled dogs										
Quarantine	112	44 (39.2)	20 (17.9)	28 (25.0)	11 (9.8)	6 (5.4)	2 (1.8)	2 (1.8)	0 (0.0)	1 (0.9)
Pound	83	39 (46.9)	29 (34.9)	10 (12.1)	4 (4.8)	16 (19.3)	10 (12.1)	2 (2.4)	0 (0.0)	0 (0.0)
TTSPCA <sup>d</sup>	70	34 (48.6)	25 (35.7)	10 (14.3)	7 (10.0)	12 (17.1)	7 (10.0)	2 (2.9)	5 (7.1)	0 (0.0)
Total	265	117 (44.2)	74 (27.9)	48 (18.1)	22 (8.3)	34 (12.8)	19 (7.2)	6 (2.3)	5 (1.9)	1 (0.4)
Veterinary establishment dogs										
Hospitals <sup>e</sup>	99	74 (74.7)	39 (39.4)	58 (58.6)	34 (34.3)	16 (16.2)	9 (9.1)	8 (8.1)	6 (6.1)	1 (1.0)
Clinics <sup>f</sup>	53	27 (50.9)	15 (28.3)	14 (26.4)	10 (18.9)	9 (16.9)	9 (16.9)	1 (1.9)	2 (3.8)	1 (1.9)
Total	152	101 (66.5)	54 (35.5)	72 (47.4)	44 (28.9)	25 (16.5)	18 (11.8)	9 (5.9)	8 (5.3)	2 (1.3)

<sup>a</sup>Resistant to one or more antimicrobial agents, <sup>b</sup>KF - Cephalothin, AMP - Ampicillin, SXT - Sulphamethoxazole/Trimethoprim, K - Kanamycin, N - Neomycin, NA - Nalidixic acid, CN - Gentamicin, Nor - Norfloxacin, <sup>c</sup>Includes dogs from households, dairy farms and hunting, <sup>d</sup>Trinidad and Tobago Society for the Prevention of Cruelty to Animals, <sup>e</sup>In-patients, <sup>f</sup>Out-patients

( $P > 0.05$ ;  $\chi^2$ ) was observed in the prevalence of resistance for *E. coli* isolated from the various sources, with prevalence of 39.2%, 46.9% and 48.6% observed for dogs kept in quarantine, pound and TTSPCA respectively.

For veterinary establishments however (Table 2), *E. coli* isolates from hospitalized dogs (74.7%) had a significantly ( $P < 0.05$ ;  $\chi^2$ ) higher prevalence of resistance than that observed for *E. coli* isolates from the clinic dogs (50.9%). Overall, the prevalence of resistance was highest for ampicillin (47.4%) compared with the other antimicrobial agents tested.

Table 3. depicts the prevalence of dogs positive for resistant *E. coli* isolates from selected sources (households, dairy farms, hunting dogs) according to veterinary services

Table 3. Prevalence of *E. coli* isolates showing antibiotic resistance amongst dogs from selected sources according to veterinary services received

Source of samples <sup>a</sup>	Veterinary services					
	Received			Not received		
	N <sup>o</sup> of dogs	N <sup>o</sup> of <i>E. coli</i> isolates tested for antibiotic resistance	N <sup>o</sup> (%) of <i>E. coli</i> isolates tested positive for antibiotic resistance	N <sup>o</sup> of dogs	N <sup>o</sup> of <i>E. coli</i> isolates tested for antibiotic resistance	N <sup>o</sup> (%) of <i>E. coli</i> isolates tested positive for antibiotic resistance
Households	209	70	33 (47.1)	141	52	23 (44.2)
Dairy farms <sup>b</sup>	55	22	6 (27.3)	55	34	7 (20.6)
Hunting dogs	94	44	20 (51.3)	6	3	2 (66.7)
Total	358	136	59 (43.4)	202	89	32 (35.9)

<sup>a</sup>Dogs from clinics and hospitals which routinely received veterinary services were excluded from the source of samples, <sup>b</sup>Include Waller field (25) and Carlsen field (25)

Table 4. Comparison of antibiograms of virulence marker-negative and virulence marker-positive *E. coli* isolates

Status of <i>E. coli</i>	N <sup>o</sup> isolates tested <sup>a</sup>	N <sup>o</sup> (%) of isolates resistant <sup>b</sup>
Virulence marker-negative	125	30 (24.0)
Virulence marker-positive <sup>c</sup>	454 <sup>d</sup>	262 (57.7)
Total	579	292 (50.4)

<sup>a</sup>Sources of *E. coli* include dogs from the quarantine station, households, hospitals, pound, TTSPCA, dairy farms, clinics, hunting dogs and pet shops; <sup>b</sup>Resistant to one or more antimicrobial agents; <sup>c</sup>Virulence markers assayed for include: mucoid (100), Haemolytic (81), non-sorbitol fermenters (133), 'enteropathogenic' (187), verocytotoxigenic (74); <sup>d</sup>Some isolates were positive for more than one virulence marker

received. From 358 dogs which received veterinary services, of a total of 136 *E. coli* isolates tested, 59 (43.4%) were resistant to antimicrobial agents. However, for the 202 dogs that did not receive veterinary services, of 89 *E. coli* isolates tested, 32 (35.9%) exhibited resistance to antimicrobial agents. The difference was not statistically significant ( $P>0.05$ ;  $\chi^2$ ).

A comparison of the prevalence of the antibiotic resistance amongst virulence marker-positive and virulence marker-negative isolates of *E. coli* from all sources is shown in Table 4. Overall, 292 (50.4%) of 579 *E. coli* isolates were resistant to one or more antimicrobial agents. Of a total of 125 virulence marker-negative *E. coli* isolates tested, 30 (24.0%) were resistant, a prevalence significantly ( $P<0.001$ ;  $\chi^2$ ) lower than detected amongst virulence marker-positive isolates, 57.7% (262 of 454).

A total of 45 resistance patterns were observed amongst 309 isolates of *E. coli* from dogs from all sources which exhibited resistance to one or more antimicrobial agents. Overall, the predominant resistance patterns were KF (25.6%), AMP (13.3%) and KF-AMP (10.0%). Among the 56 *E. coli* isolates from household dogs which exhibited resistance, a total of 16 different resistance patterns were observed. The predominant resistance patterns were KF (42.9%), KF-K (14.3%) and KF-AMP (7.1%).

For kennelled dogs, amongst 44 resistant isolates of *E. coli* isolated from quarantine dogs, a total of 14 resistance patterns were observed with the predominant resistance patterns being AMP (34.1%), KF-AMP (18.2%) and KF (13.6%).

Of the 39 resistant *E. coli* isolates recovered from pound dogs, 15 different resistance patterns were observed with cephalothin (38.5%) being the predominant pattern.

For dogs from the TTSPCA, of the 34 resistant *E. coli* isolates exhibited, 13 different resistant patterns were detected. The predominant resistance patterns were KF (32.4%), KF-K (11.8%) and CN (11.8%).

Amongst 74 resistant *E. coli* isolates from hospitalized dogs tested, 24 resistant patterns were observed with 15 (62.5%) of these patterns consisting of resistance to three or more antimicrobial agents. The predominant resistance patterns were AMP (18.9%), KF-AMP (13.5%) and KF-SXT-AMP (10.8%). For 27 resistant *E. coli* isolates recovered from clinic dogs, 15 resistant patterns were detected with the predominant resistance patterns being KF (14.8%), KF-AMP (11.1%) and SXT (11.1%).

Only five resistance patterns were observed amongst 13 resistant canine *E. coli* isolates from dairy farms, and the most frequent pattern was KF (69.2%). Amongst 22 resistant isolates from hunting dogs, a total of 11 resistant patterns were detected. The predominant patterns were AMP (27.3%), KF (13.6%) and KF-K-N (13.6%).

## Discussion

Overall, 47.9% of *E. coli* isolates recovered from dogs in this study exhibited resistance to antimicrobial agents with a high prevalence of resistance to cephalothin (30.1%) and least resistance to norfloxacin (0.5%). It was not unexpected that resistance was least exhibited to norfloxacin as it is a relatively new drug and it was reported by GYLES (1993) that a high percentage of *E. coli* isolates are likely to be sensitive to fluoroquinolones. Although the new fluoroquinolones are less prone to develop resistance (SEMJÉN, 2000), the emergence of resistance to this group of drugs among canine *E. coli* isolates should be a source of concern.

Furthermore, the findings in the present study revealed high percentages of *E. coli* isolates being resistant to ampicillin (58.6%) and SXT (34.3%) amongst hospitalized dogs, and 6 (35.3%) of the 17 isolates that were resistant to gentamicin were also isolated from hospitalized dogs. This is not a surprise as these are three commonly used antimicrobial agents in veterinary practices in Trinidad. *E. coli* isolated from dogs presented at walk-in clinics exhibited resistance prevalence of 26.4%, 18.9% and 3.8% to ampicillin, SXT and gentamicin respectively. An earlier study conducted by ADESIYUN et al. (1997) on *E. coli* isolates from dogs sampled at a clinic revealed resistance of 42.9%, 10.2% and 24.5% to the same three antimicrobial agents in their respective order. Of the three agents, resistance to SXT was higher in the present study compared with the resistance to SXT in the earlier study (ADESIYUN et al., 1997).

The prevalence of resistance of *E. coli* isolates to antimicrobial agents was significantly different across the various sources of dogs sampled, ranging from 23.2% in dairy farm dogs to 74.7% in hospital dogs. Hospitalized dogs which are housed in an environment that selects for antimicrobial resistance due to a widespread use of antimicrobial agents have been reported to develop resistance (ANONYMOUS, 1998; NORMAND et al., 2000). Additionally, hospitalized dogs undergo therapeutic care, prophylaxis and chemotherapy whereas dairy farm dogs very rarely receive antibiotic treatment, which may be responsible for the low prevalence of resistance detected in this group of dogs.

The rather high number of 45 resistance patterns detected amongst *E. coli* isolates from dogs from all sources with predominant resistance patterns being KF (25.6%), AMP (13.3%) and KF-AMP (10.0%), is a reflection of the generally high levels of resistance to antimicrobial agents. Resistance patterns may demonstrate multiple resistance to many antimicrobial agents and could have therapeutic consequences.

The detection of 24 different resistance patterns in hospitals with over 60% of the patterns exhibiting multi-resistance was also not unexpected. Resistance to antimicrobial agents is most common in areas with high usage of antibiotics such as hospitals (STERNBUERG, 1999). Additionally, the widespread use of antimicrobial agents and

the existence of multi-resistant gram-negative bacteria in hospital settings have been described previously (FINLAND, 1972; MURRAY and MOELLERING, 1987).

The predominant resistance pattern, AMP (18.9%), observed in hospitalized dogs, is comparable with the predominance of AMP observed for hunting dogs (27.3%) and quarantine dogs (34.1%), an indication of a widespread usage of this antimicrobial agent in dogs from these sources. Resistance to ampicillin however, was observed to feature in the resistance patterns of isolates from dogs from all the other sources. This suggests ampicillin usage alone may not explain the resistance exhibited by *E. coli* isolates from various sources in the environment.

The presence of 15 different resistance patterns in pound dogs with 8 patterns inclusive of ampicillin, may be a reflection of exposure to antimicrobial agents that may exist in the environment, as these dogs are not seen by veterinarians. LINTON (1986) noted that carriage of resistant isolates in animals can be due to ingestion of resistant organism from the environment.

Although there can be no doubt that antibiotic resistance in animals could be a result of veterinary administration of antimicrobial agents (SMITH and LEWIN, 1993), resistance can also be observed in animals that receive no antibiotics (FINLAND, 1972). This is in agreement with the findings in the present study where a similar prevalence (43.4%) of antibiotic resistance was observed for *E. coli* isolates from dogs that received veterinary services compared with the *E. coli* isolates in dogs that did not receive veterinary services (35.9%).

It has been established that animals, particularly apparently healthy ones, may be sources of resistant isolates of *E. coli* and other pathogens from animals to man (SMITH and LEWIN, 1993). DAVIES and STEWART (1978) have confirmed this interrelationship between domestic pets and human beings through the spread of R-plasmids while MONAGHAN et al. (1981) isolated bacteria with transferable antimicrobial resistant determinants from the rectums of healthy dogs and cats. Antimicrobial resistance in companion animals would be potentially important because pets are present in the home and have close contact with humans. Also, the spread of resistant isolates of *E. coli* from hospital dogs and cats to other in-patients, as well as to the general population outside, should be considered as a source of concern.

The prevalence of resistance amongst virulence marker-positive isolates of *E. coli* was significantly higher than found amongst virulence marker-negative isolates from pet animals. This is of public health significance as virulence marker-positive isolates causing infection or disease and being resistant to antimicrobial agents would have dire consequences.

It was concluded that the relatively high prevalence of resistance to antimicrobial agents amongst *E. coli* isolates from non-diarrhoeic pet animals in Trinidad was high and may pose zoonotic and therapeutic problems.

---

### Acknowledgements

We would like to thank the Pan American Health Organization (PAHO) for funding the project. We also express our gratitude to M. Negleka, D. Simeon, the U.W.I Campus Research Committee, the staff at the Quarantine station, TTSPCA, the Dog Pound, Vet Hospitals and Clinics, Pet shops and the various pet owners throughout Trinidad for their assistance. We also acknowledge the contributions of G. Ramirez, E. Neptune and C. Gall.

### References

- ADESIYUN, A. A., M. CAMPBELL, J. S. KAMINJOLO (1997): Prevalence of bacterial enteropathogens in pet dogs in Trinidad. *J. Vet. Med. B* 44, 19-27.
- ANONYMOUS (1998): Standing Medical Advisory Committee, Sub-group on Antimicrobial resistance. The path of least resistance. Department of Health, UK.
- BAUER, A. W., W. M. M., KIRBY, J. C. SHERRIS, M. TURCK (1966): Antibiotic susceptibility testing by a standardized single disk method. *Techn. Bull. Regist. Med. Technol.* 36, 493-496.
- DAVIES, J. E. (1998): Origins, acquisition and dissemination of antibiotic resistance determinants. In: *Antibiotic Resistance: Origin, Evolution, Selection and Spread: Ciba Foundation Symposium 207*, Wiley, Chichester; p. 15-27.
- DAVIES, M., P. R. STEWART (1978): Transferable drug resistance in man and animals: genetic relationship between R-plasmids in enteric bacteria from man and domestic pets. *Aust. Vet. J.* 54, 507-512.
- DECKER, M. D., W. SCHAFFNER (1992): The relationship between the hospital and the community. In: *Hospital Infections* (Bennett, J.V., P. S. Brachman, Eds.). 3<sup>rd</sup> ed. Boston, Little, Brown and Company. pp. 221-230.
- ESPINASSE, J. (1993): Responsible use of antimicrobials in veterinary medicine: perspectives in France. *Vet. Microbiol.* 35, 289-301.
- FINLAND, M. (1972): Changing patterns of susceptibility of common bacterial pathogens to antimicrobial agents. *Ann. Intern. Med.* 76, 1009-1036.
- GYLES, C. L. (1993): *Escherichia coli*. In: *Pathogenesis of bacterial infections in animals*. (Gyles C.L., C. O. Thoen, Eds.), 2<sup>nd</sup> ed. Iowa: Iowa State University Press. pp. 164-187.
- HELMUTH, R. (2000): Antibiotic resistance in *Salmonella*. In: *Salmonella in Domestic Animals*, (Wray, C., A. Wray, Eds.) CAB International. pp. 89-106.
- KONOWALCHUK, J., J. I. SPEIRS, S. STAVRIC (1977): Veroreponse to a cytotoxin of *Escherichia coli*. *Infect. Immun.* 18, 775-779.
- LEVY, S. B. (1998): The challenge of antibiotic resistance. *Sci. Am.* 278, 46-53.
- LINTON, A. H. (1986): Flow of resistance genes in the environment from animals to man. *J. Antimicrobial. Chemother.* 18, 189-197.

N. Seepersadsingh and A. A. Adesiyun: Pathogenicity and antibiotic sensitivity of *Escherichia coli* isolates from non-diarrhoeic dogs

- MACFADDIN, J. F. (1980): Biochemical tests for identification of bacteria. Williams and Wilkins, New York.
- MARCH, S. B., S. RATNAM (1986): Sorbitol-MacConkey medium for detection of *Escherichia coli* O157:H7 associated with haemorrhagic colitis. J. Clin. Microbiol. 23, 869-872.
- MONAGHAN, C., U. TIERNEY, E. COLLERAN (1981): Antibiotic resistance and R-factors in the faecal coliform flora of urban and rural dogs. Antimicrob. Agents Chemother. 19, 266-270.
- MURRAY, B. E., R. C. MOELLERING Jr. (1987): Patterns and mechanisms of antibiotic resistance. Med. Clin. North. Amer. 62, 899-923.
- NORMAND, E. H., N. R. GIBSON, S. W. J. REID, S. CARMICHEAL, D. J. TAYLOR (2000): Antimicrobial-resistance trends in bacterial isolates from companion-animal community practice in the UK. Prev. Vet. Med. 46, 267-278.
- ØRSHOV, I., F. ØRSHOV (1984): Serotyping of *Escherichia coli*. In: Methods in Microbiology. Vol 14, (Bergan T., Ed.) London, Academic Press. pp. 43-112.
- SEMJÉN, G. (2000): The effects of intervention on antimicrobial resistance. In: Proceedings of the symposium on antibiotic resistance with emphasis on animal-human transfer. 13-14 Sept. 1999. Falken, Sweden. Acta Vet. Scand., Suppl. 93, 105-110.
- SMITH, J. T., C. S. LEWIN (1993): Mechanisms of antimicrobial resistance and implications for epidemiology. Vet. Microbiol. 35, 233-242.
- STERNBUERG, S. (1999): Antimicrobial resistance in bacteria in pets and horses Acta. Vet. Scand. Suppl. 92, 37-50.

Received: 6 February 2006

Accepted: 2 November 2007

---

**SEEPERSADSINGH, N., A. A. ADESIYUN: Patogenost izolata bakterije *Escherichia coli* iz pasa bez proljeva i njihova osjetljivost prema antibioticima. Vet. arhiv 77, 531-542, 2007.**

**SAŽETAK**

Izolati *E. coli* iz pasa u Trinidadu bili su testirani na osjetljivost prema antimikrobnim tvarima difuzijskim testom. Upotrijebljene su sljedeće antimikrobne tvari i njihove koncentracije: cefalotin (KF, 30 µg), ampicilin (AMP, 10 µg), kanamicin (K, 30 µg), neomicin (N, 30 µg), gentamicin (CN, 10 µg), sulfametoksazol/trimetoprim (SXT, 23.25/1.75 µg), nalidiksična kiselina (NA, 30 µg) i norfloksacin (NOR, 10 µg). Sveukupna prevalencija otpornosti na jednu ili više antimikrobnih tvari za izolate *E. coli* iz pasa iznosila je 47,9%. Razlika u prevalenciji izolata s različitih izvora bila je statistički značajna ( $P < 0,001$ ;  $\chi^2$ ). Općenito je otpornost bila veća prema cefalotinu (30,1%). Ukupno je 45 izolata pokazivalo otpornost s pretežitošću prema cefalotinu (25,6%). Zaključeno je da relativno velika prevalencija izolata *E. coli* iz pasa bez proljeva u Trinidadu otpornih prema antimikrobnim tvarima može predstavljati problem u zoonotskom i terapijskom smislu.

**Cljučne riječi:** antimikrobna osjetljivost, *Escherichia coli*, pas

---