UNIVERSITI PUTRA MALAYSIA

OCCURRENCE OF ANTIBIOTIC RESISTANT *Salmonella* spp. IN STRAY AND OWNED CATS

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OCCURRENCE OF ANTIBIOTIC RESISTANT *Salmonella* spp. IN STRAY AND OWNED CATS

NUR FARAWAHIDAH BINTI MOHSIN

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It is hereby certified that I have read this project paper entitled “Occurrence Of Antibiotic Antibiotic Resistant *Salmonella* Spp. In Stray and Owned Cats”, by Nur Farawahidah Binti Mohsin and in my opinion it is satisfactory in terms of scope, quality and presentation as partial fulfilment of the requirement for the course VPD 4999 – Final Year Project.

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To

My beloved mother and father,

Mohsin bin Mudakir and Norlidah Ahmad,

My lovely family,

My bestfriends,

My coursemates,

Friends.

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Alhamdulillah,

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ABSTRACT

An abstract of the project paper presented to the Faculty of Veterinary Medicine in partial fulfilment of the course VPD 4999 – Final Year Project

OCCURRENCE OF ANTIBIOTIC RESISTANT *Salmonella* spp. IN STRAY AND OWNED CATS

By

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2016

Supervisor: Prof. Dr. Saleha Bt. Abdul Aziz

Salmonellosis is an important zoonotic disease worldwide. The organisms reside commonly in the gastrointestinal tracts. Cats are most widely kept pet animals, yet the risk that these animals pose for transmission of *Salmonella* to humans is unclear. Free roamer and stray cats are potential candidates for *Salmonella* carriage and might contribute actively to the contamination of environment. The objectives of this study were to determine the occurrence of *Salmonella* spp. and to determine the antimicrobial resistance of *Salmonella* spp. in stray and owned cats. A total of 60 rectal

swab samples were collected which consisted of 30 from owned cats at veterinary clinics and individual owners and the other 30 samples were taken from stray cats at residential areas and food stalls. Out of 60 cats, only three (5%) were positive for *Salmonella* spp. which were from stray cats (10%). The *Salmonella* spp. isolated were tested against six different antibiotics namely ampicillin, chloramphenicol, ciprofloxacin, gentamicin, sulfamethoxazole-trimethoprim and nalidixic acid. All three (100%) isolates were resistant to ampicillin, but susceptible to ciprofloxacin, and one (33%) isolate was susceptible to gentamicin and chloramphenicol. This could be expected as stray cats are often exposed to intestinal infection with *Salmonella* species than pet household cats. Stray cats may pose a potential threat to public health and their faecal materials may play significant roles in the contamination of environment.

Keywords: Stray cats, owned cats, *Salmonella* spp., antibiotic resistant

ABSTRAK

Abstrak daripada kertas projek yang dikemukakan kepada Fakulti Perubatan Veterinar sebagai memenuhi sebahagian daripada kursus VPD 4999 – Projek Ilmiah Tahun Akhir.

KEHADIRAN *Salmonella* spp. TAHAN ANTIBIOTIK DIASINGKAN DARIPADA KUCING LIAR DAN PELIHARAAN ORANG

Oleh

Nur Farawahidah Binti Mohsin

2016

Penyelia: Prof. Dr. Saleha Bt. Abdul Aziz

Salmonellosis adalah penyakit zoonotik penting di seluruh dunia. Organisma ini biasanya tinggal dalam saluran pencernaan. Kucing adalah haiwan yang paling banyak dipelihara sebagai haiwan kesayangan, namun risiko haiwan ini dalam pemindahan *Salmonella* kepada manusia adalah tidak jelas. Kucing 'free-roamer' dan kucing liar berpotensi menyebarkan *Salmonella* dan menyumbang secara aktif kepada pencemaran alam sekitar. Objektif kajian ini adalah untuk menentukan kehadiran *Salmonella* spp. dan untuk menentukan rintangan antimikrob *Salmonella* spp. dalam

kucing liar dan kucing peliharaan orang. Sebanyak 60 sampel swab rektum dikumpulkan yang terdiri daripada 30 dari kucing peliharaan di klinik veterinar dan pemilik individu dan 30 sampel selebihnya diambil dari kucing liar di kawasan perumahan dan gerai-gerai makanan. Daripada 60 kucing, hanya tiga (5%) didapati positif *Salmonella* spp. yang juga merupakan daripada kucing liar (10%). *Salmonella* spp. yang diasingkan telah diuji terhadap enam antibiotik yang berbeza iaitu ampicillin, chloramphenicol, ciprofloxacin, gentamicin, sulfamethoxazole-trimethoprim dan nalidixic acid. Ketiga-tiga (100%) sampel menunjukkan ketahanan terhadap antibiotik ampicillin, tetapi sensitif kepada ciprofloxacin, dan satu (33%) sampel sensitif kepada gentamicin dan chloramphenicol. Ini boleh dijangka kerana kucing liar seringkali terdedah kepada jangkitan usus dengan spesies *Salmonella* berbanding kucing dipelihara orang. Kucing liar boleh menimbulkan ancaman yang berpotensi kepada kesihatan awam dan bahan-bahan najis mereka boleh memainkan peranan penting dalam pencemaran alam sekitar.

Kata kunci: kucing liar, kucing dimiliki, *Salmonella* spp, tahan antibiotik.

1.0 INTRODUCTION

Salmonellosis has long been recognized as an important zoonotic disease of worldwide economic significance. *Salmonella* genus is a member of the Enterobacteriaceae family, comprising Gram-negative rod-shaped nonspore-forming bacteria. The organisms inhabit the intestinal tracts of vertebrate and invertebrate animals worldwide and its excretion results in contamination of environment, feed, water and infected animals (Seepersadsingh *et al*., 2005; McDonough, 2000). Disease symptoms in human include acute abdominal pain, diarrhoea, nausea, fever, and sometimes vomiting. Mild symptoms are often seen and the infection can also occur without symptoms.

Considering the high frequency of food contamination and the emergence of multidrug−resistant *Salmonella* strains, control of *Salmonella* in food-producing animals has become a worldwide challenge. The role of pet animals as a source of *Salmonella* has not been fully investigated, but severe human infections originating from reptiles, especially pet turtles, have been reported (Immerseel *et al*., 2004). Cats and dogs are the most widely kept pet animals, yet the incidence of *Salmonella* in these animals is largely unknown, and the risk that these animals pose for transmission of *Salmonella* to humans is unclear. In particular, cats that can freely roam outside, and are able to scavenge or hunt food are potential candidates for *Salmonella* carriage (Immerseel *et al*., 2004).

Clinical salmonellosis in cats is relatively uncommon and few references on it exist in scientific literature (McDonough, 2000). Cats appear to be highly resistant to *Salmonella* infection unless they are stressed by overcrowding, dietary changes, transport, hospitalization, antimicrobial therapy, or concurrent illness at the time of *Salmonella* exposure (McDonough, 2000). Most reports concerning *Salmonella* and cats are case studies of clinical salmonellosis, which result in septicaemia and death. Subclinical infections and carrier animals, however, are more important with respect to transmission to humans (Immerseel *et al*., 2004). Since, having cats living closely at home has become more common in Malaysia, epidemiological data of *Salmonella* and antimicrobial resistance patterns are needed in order to prevent and control *Salmonella* spp. in cats. Thus, the objectives of this present study were:

1) to study the occurrence of *Salmonella* in cats from owned and stray cats.
2) to compare the isolation rate of *Salmonella* between owned and stray cats.
3) to determine antibiotic resistance level of *Salmonella* spp. isolated.

2.0 LITERATURE REVIEW

2.1 *Salmonella* spp.

Salmonella are gram-negative, rod-shaped, non-spore forming, and predominantly motile by means of peritrichous flagella with diameters of around 0.7-1.5µm and lengths of 2-5µm (Public Health England, 2015).

According to Public Health England (2015), all *Salmonella* serotypes are considered pathogenic. Some serotypes are host-specific, but the majority can affect different hosts. Most *Salmonella* serotypes possess two phases of H (flagellar) antigens. *Salmonella enteritidis* and *Salmonella typhimurium* are two dominant serotypes transmitted from animals to humans in most part of world. *Salmonella typhi* and *Salmonella paratyphi* A, B and C are the most common causes of enteric fever in humans. *Salmonella* serovars of *Salmonella enterica* are found in faeces, blood, bile, urine, food and feed and also environmental materials.

2.2 Prevalence of *Salmonella* spp. in cats

Salmonellosis in cats is rare and is believed to precipitate by stressed such as overcrowding, dietary changes, transport, hospitalization, antimicrobial therapy, or concurrent illness at the time of *Salmonella* exposure. According to Seepersadsingh *et al*. (2005), *Salmonella* carrier state in most cats is clinically inapparent and prevalences of carriers were found variable. The isolation rates of *Salmonella* serovars ranged from 0.0% to 14.0% (Seepersadsingh *et al*., 2005).

Bhaiyat *et al*. (2009) reported in a case of concurrent lymphosarcoma and *Salmonella enteritidis* infection in a cat suggested that *Salmonella* can cause generalized infection in cats with lyphosarcoma; however, not all cats with lymphoma may develop salmonellosis. Another study reported in Thailand, *Salmonella* isolation from diarrheal and non-diarrheal cats showing 8.0% and 10.0% were infected with *Salmonella* respectively (Polpakdee *et al.*, 2012). Cats have high intrinsic resistance to salmonellosis, therefore, asymptomatic carrier state, and subclinical disease are more common in adult cats (Bhaiyat *et al.*, 2009).

In a study on the prevalence of enteric zoonotic organisms in cats found that a higher proportion of shelter cats (18.2%) compared with client-owned cats (10.1%) had enteric organisms (Hill *et al*., 2000). According to Immerseel *et al*. (2004), in other study on cats as the risk for transmission of antimicrobial drug-resistant *Salmonella*, it was found that grouped-housed cats (51.4%) had highest proportion of *Salmonella* isolation, followed by diseased cats (8.6%) and healthy house cats (0.36%). Stray cats and shelter cats are more likely to excrete Salmonella in their faeces than pet household cats (Seepersadsingh *et al*., 2005).

2.3 Antimicrobial Usage and Antimicrobial Resistance Microorganism in Pet Animals

Widespread use of broad-spectrum antimicrobials has been reported in small animal practice in Europe (ESVA, 2012). The most commonly used antimicrobials for dogs and cats in Denmark, Finland, Italy, Sweden, Norway and UK are beta lactams

such as amoxicillin and amoxicillin combined with clavulanic acid (EMA, 2013). Lincosamides (clindamycin), fluoroquinolones, macrolides, tetracyclines (doxycycline), nitroimidazoles and trimethoprim-sulfonamides have also been reported to be routinely used in small animal practice but on a smaller scale than beta-lactams (EMA, 2013).

Lack of confirmed diagnosis could lead to the misuse of antimicrobials. Findings from a study in Italy revealed that only 5% of antimicrobial prescriptions in a veterinary teaching hospital were supported by results of culture and antibiotic sensitivity test (Escher *et al*., 2011). Antimicrobials administration has been reported to treat acute diarrhoea in dogs (German *et al*., 2010) and feline lower urinary tract disease (Thomson *et al*., 2009), for which conditions antimicrobial treatment is usually not recommended (Guardabassi *et al*., 2008). A cross-sectional study on antimicrobial prescribing patterns in the UK showed that approximately 2% of prescriptions for dogs and cats were for products not authorised in those species (Hughes *et al*., 2012).

Increasing antimicrobial resistance among *Enterobacteriaceae* is emerging as a significant public health concern in human medicine (Pitout and Laupland, 2008). Multidrug-resistant *Salmonella* Typhimurium have been associated with outbreaks of gastrointestinal nosocomial infections in companion animals in veterinary clinics and an animal shelter (Wright *et al*., 2005). One such outbreak also involved veterinary staff and other persons who were in contact with animals (Cherry *et al*., 2004). Companion animal facilities may serve as foci of transmission for salmonellae between animals and humans if adequate infection control measures are not followed (Wright *et al*., 2005).

2.4 Public Health Significance

Salmonella infections in man are usually acquired by eating contaminated food; however, direct contact with infected animals can result in exposure and infection of *Salmonella*. The role of pet animals as a source of *Salmonella* has not been fully investigated, but severe human infections originated from reptiles, especially pet turtles, have been reported (Immerseel *et al.*, 2004). According to Mirmomeni *et al*. (2009), *Salmonella enterica* subspecies *enterica* (subspecies I) is responsible for 99.5% of the infections in human and animals. *Salmonella enteriditis* is a major cause of human food-borne illness and the most frequent serovar detection in outbreaks of human salmonellosis.

A unique and critical aspect related to antimicrobial resistance in companion animals is their close contact with humans. This creates opportunities for interspecies transmission of (multidrug) resistant bacteria. Use of antimicrobials that are critically important for human health in companion animals is an additional risk factor for emergence and transmission of antimicrobial resistance. Yet, the current knowledge of many aspects of this field is limited and no assessment of this specific risk is performed when approving new veterinary antimicrobials. Cats that freely roam outside are therefore able to scavenge or hunt food are potential candidates for *Salmonella* carriage (Immerseel *et al*., 2004).

3.0 MATERIALS AND METHODS

3.1 Study Design

A prevalence study was conducted from 9th January to 14th February 2016. In this study, 60 stray and owned cats were randomly selected and sampled for the presence of *Salmonella* spp.

3.2 Sources of samples

A total of 60 rectal swab samples were collected which consisted of 30 from owned cats at veterinary clinics and individual owners and the other 30 samples were taken from stray cats at residential areas and food stalls. The target populations were stray cats at Serdang and owned cats from veterinary clinics around Klang Valley, from which rectal swab samples were taken. The samplings were done in two (2) weeks with an average of 30 cats sampled per week. Thirty owned cats were chosen randomly from walk-in owners who brought for veterinary service or from resident cats in that veterinary clinic. Consent and relevant information regarding the pet's signalment and health status were taken from owner before rectal swab was taken.

3.3 Samples collection

The cats were restraint with the help of the owner or the veterinarian in charge. Each rectal swab was obtained by inserting the moistened sterile cotton swab into the rectal region of a cat as deep as 1 inch and rotated gently before taken out.

3.4 Samples storage

Individual rectal swab was placed into a separate universal bottle containing 10 ml of Buffered Peptone Water (BPW) as pre-enrichment broth. The pre-enrichment broth containing the samples were transported in cool ice box.

3.5 Isolation of *Salmonella* spp.

The samples were bought to Veterinary Public Health Laboratory in Faculty of Veterinary Medicine, Universiti Putra Malaysia for isolation and identification of *Salmonella* spp..

The cotton swabs in a universal bottles filled with BPW were incubated at 37 °C for 24 hours as pre-enrichment stage. Then, 0.1 ml of each BPW culture was transferred into the bottle filled with 10 ml of Rappaport-Vassiliadis (RV) enrichment broth and was incubated at 42 °C for 24 to 48 hours as enrichment stage. A loopful of the culture from RV enrichment broth was streaked onto Xylose Lysine Deoxycholate (XLD) agar and Brilliant Green Agar (BGA) before intubated at 37 °C for 24 to 48 hours. Presumptive *Salmonella* spp. colonies appeared as black or black centred colonies on red-violet background on XLD

agar, while appeared as red to pink-white colonies surrounded by a red zone on BGA agar. At least three colonies were selected from each plate and subculture was done on XLD agar in order to obtain pure culture for biochemical tests.

3.6 *Salmonella* spp. Identification Test

Pure colonies suspected for *Salmonella* spp. were subjected to standard biochemical tests for identification.

Biochemical tests included were:

i. Sulphide Indole Motility Test (SIM) – one colony from pure culture was picked with flamed straight wire and stabbed directly into the agar. The test tube was incubated at 37 °C for 24 hours. After incubation period, indole test was confirmed by dropping few drops of Kovacs' Reagent into the test tube.

ii. Triple Sugar Iron Test (TSI) – one colony from pure culture was picked with flamed straight wire and stabbed directly into the agar slant and the surface streaked in zig-zag manner. The test tube was incubated at 37 °C for 24 hours.

iii. Urease Test - one colony from pure culture was picked with flamed straight wire and the surface was streaked in zig-zag manner. The test tube was incubated at 37 °C for 18 to 24 hours.

iv. Citrate Test - one colony from pure culture was picked with flamed straight wire and the surface was streaked in zig-zag manner. The test tube was incubated at 37 °C for 24 to 48 hours.

v. Lysine Iron Agar Test (LIA) - one colony from pure culture was picked with flamed straight wire and stabbed directly into the agar slant and the surface will be streaked in zig-zag manner. The test tube was incubated at 37 °C for 24 hours.

Cultures showing typical reactions of *Salmonella* spp. on TSI showed acid butt (yellow), alkaline slant (red) with hydrogen sulphate (black) and gas production. On LIA, the result showed Lysine decarboxylase (LDC) positive butt and slant (violet) with hydrogen sulphate (black), while on SIM showed positive motilility with or without hydrogen sulphate positive (black), positive (blue) on citrate test and negative (yellow) on urease test.

Colonies showing typical reactions of *Salmonella* spp. from biochemical tests were then subjected to Slide Agglutination Test (SAT) using *Salmonella* polyvalent 'O' and 'H' Antisera A-S. A drop of normal saline was put on glass slide. Then, a loopful of presumptive *Salmonella* colonies was taken and mixed well with the normal saline on glass slide to produce dense suspension. Then, a drop of *Salmonella* polyvalent antiserum was dropped onto the suspension and mixed well. The slide was held in between fingers and gently rocked back and forth for at least 1 minute. If distinct agglutination occurred, the isolate was regarded as positive as shown in Plate 6. The positive *Salmonella* spp. were cultured on Nutrient agar for antibiotic sensitivity test.

3.7 Antibiotic Sensitivity Test

Five loopfuls of *Salmonella* spp. pure culture from each nutrient agar was mixed with 4 ml Tryptic Soy Broth (TSB) and incubated at 37 °C for four to six hours. The concentration of TSB was adjusted using sterile TSB until it reached turbidity of 0.5 McFarland standard suspensions. A swab of the mixture was streaked evenly onto Mueller Hinton agar (Oxoid, UK) plate for each pure culture and left it to semi dry.

The susceptibility of the organism to antimicrobial agents was tested by standard disc diffusion method. Antimicrobial agents chosen were ampicilin (10 µg), chloramphenicol (30 µg), ciprofloxacin (5 µg), nalidixic acid (30 µg), gentamicin (10 µg), and trimethoprim-sulphamethoxazole (25 µg). The plates were incubated at 37 °C for 24 hours. Diameter of inhibition zone for each antibiotic disc was measured using calliper for determination of susceptibility towards the chosen antibiotics.

3.8 *Salmonella* spp. serotyping

The colonies confirmed for *Salmonella* spp. through the Slide Agglutination test were sent to Veterinary Research Institute (VRI) in Ipoh on nutrient agar slant for further serotyping.

4.0 RESULT

4.1 Isolation of *Salmonella* spp. from Rectal Swab of Cats

A total of 60 rectal swab samples were collected which consisted of 30 from owned cats and the other 30 samples were taken from stray cats. Twenty-eight swabs of owned cats were obtained from veterinary clinics, and two swabs of owned cats were obtained from individual owners who kept cats as pets. For stray cats, two swabs were obtained from food stalls, two swabs were from residential college, and remaining 26 swabs were from residential areas in Serdang.

Table 1: Isolation of *Salmonella* spp. in cats

Animals	Sources of samples	Number of samples collected	No. of positive samples	Percentage of samples positive (%)
Owned cats	Veterinary clinic (resident cat)	18	0	0
	Veterinary clinic (walk-in and boarding)	10	0	0
	Individual owners	2	0	0
	Total	**30**	**0**	**0**
Stray cats	Household	26	3	11.5
	College	2	0	0
	Food stalls	2	0	0
	Total	**30**	**3**	**10**
	Overall total	**60**	**3**	**5**

Out of 60 samples, only three samples from the cat was positive for *Salmonella* spp. (Table 1).

4.2 Identification of *Salmonella* spp.

Salmonella spp. was presumptively isolated from 15 samples of stray cats and three (3) samples of owned cats through colour differentiation on Xylose Lysine

Deoxycholate (XLD) agar and Brilliant Green Agar (BGA) as shown in Plate 1 and Plate 2 respectively. Out of 18 presumptive isolates from stray and owned cats, only three (5%) were identified as *Salmonella* spp. after biochemical tests (TSI, LIA, SIM, Urease and Citrate test). Upon biochemical tests: three sample showed positive result with characteristic indole negative, sulphide gas producer, urease negative, citrate negative, and positive lysine decarboxylation as shown in Plate 3. The isolate was further confirmed by using *Salmonella* 'O' Antisera and 'H' Antisera. Evidence of agglutination indicated all three were positive *Salmonella* spp.

4.3 Antibiotic Sensitivity Test

Antibiotic sensitivity test was carried out for *Salmonella* spp. as shown in Plate 4. The results are as shown in Table 2, Table 3 and Figure 1.

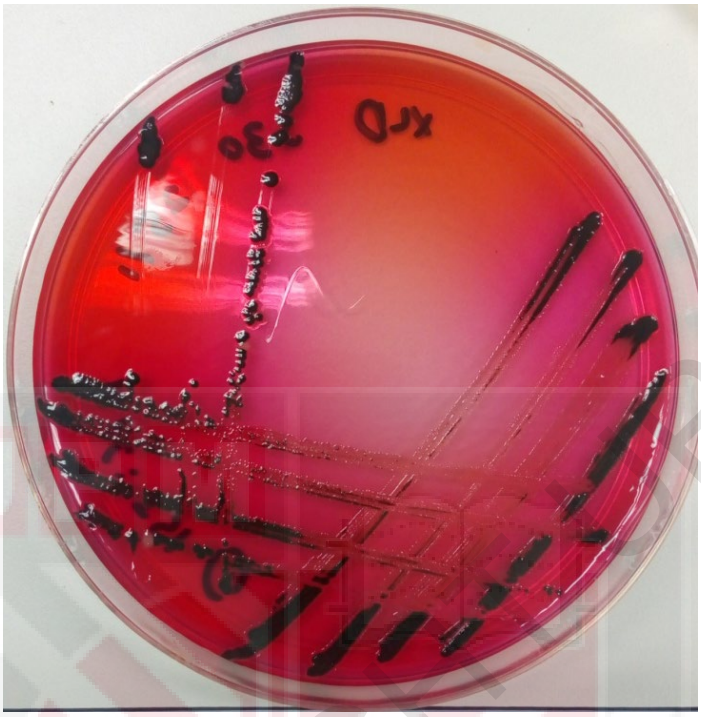


Plate 1: *Salmonella* spp. colonies on Xylose Lysine Deoxycholate (XLD) agar

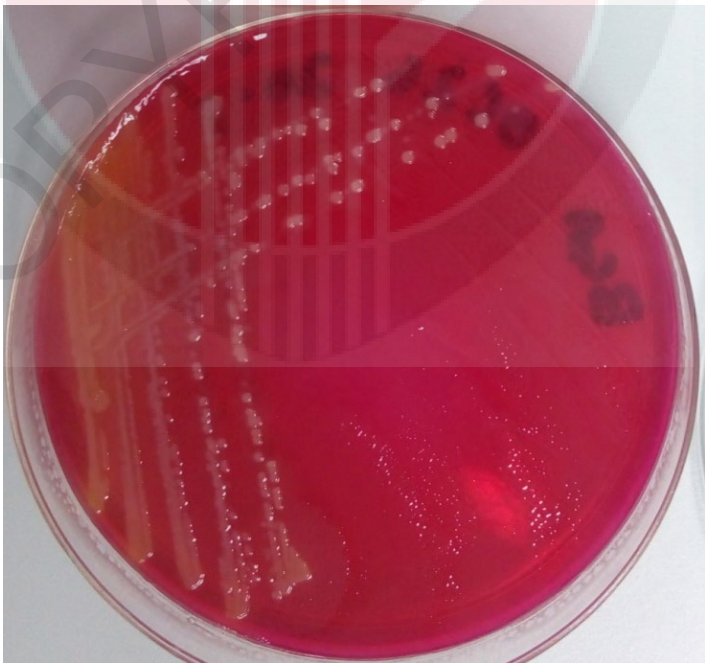


Plate 2: *Salmonella* spp. colonies on Brilliant Green Agar (BGA)

Plate 3: Positive Result on Biochemical Tests (TSI, Urease, LIA, Citrate, SIM)

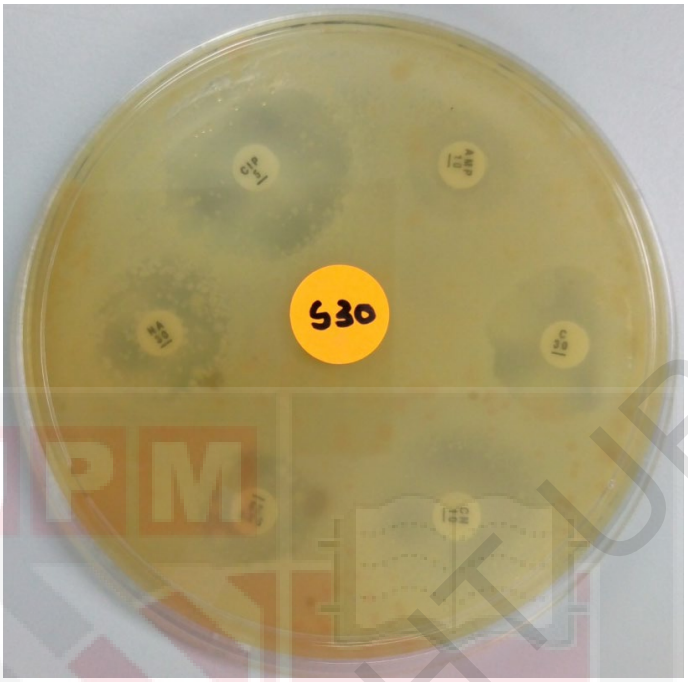


Plate 4: Antibiotic Sensitivity Test

Table 2: Measurement of Zone of Inhibition for *Salmonella* spp.

Samples	Inhibition Zone Diameter (mm)					
	A	**C**	**Cip**	**Gn**	**Sxt**	**Na**
S27	9 (R)	17 (I)	22 (S)	14 (I)	14 (I)	16 (I)
S29	10 (R)	18 (S)	22 (S)	13 (I)	12 (I)	14 (I)
S30	5 (R)	17 (I)	22 (S)	15 (S)	13 (I)	14 (I)

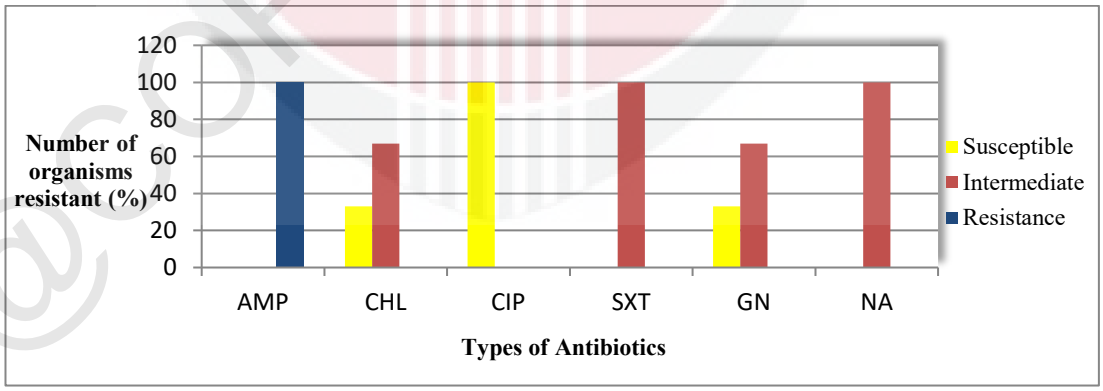
A- Ampicillin C- Chlorampenicol Cip- Ciprofloxacin Gn- Gentamicin
Sxt- Sulfamethoxazole-trimethoprim Na- Nalidixic acid

R- Resistant to antibiotic
I- Intermediate to antibiotic
S- Susceptible to antibiotic

Table 3: Susceptibilty of *Salmonella* spp. Isolates to Six Antimicrobial Agents

Antimicrobial agents	Number and Percentage resistance (%)	Number and Percentage intermediate (%)	Number and Percentage sensitivity (%)
Ampicillin	3 (100)	0	0
Chloramphenicol	0	2 (67)	1 (33)
Ciprofloxacin	0	0	3 (100)
Trimethoprim-Sulfamethoxazole	0	3 (100)	0
Gentamicin	0	2 (67)	1 (33)
Nalidixic Acid	0	100	0

Figure 1: Susceptibility of *Salmonella* spp. to six different antibiotics



Note: Ampicillin (AMP), Chloramphenicol (CHL), Ciprofloxacin (CIP), Trimethoprim-Sulfamethoxazole (SXT), Gentamicin (GN), Nalidixic acid (NA)

Of the three isolates tested for antimicrobial resistance from cats, all were resistant to ampicillin. However, all of the three isolates of *Salmonella* spp. were sensitive to ciprofloxacin, and one of the isolates was sensitive (33%) to chloramphenicol and gentamicin.

5.0 DISCUSSION

In this study, there was high occurrence of *Salmonella* in stray cats (10%) and none in owned cats. This is as expected as stray cats are often exposed to intestinal infection such as *Salmonella* spp. than pet household cats. Overall, the occurrence of 5% of *Salmonella* in cats found in current study was higher compared to 2.1% in 94 cats in Trinidad (Seepersadsingh *et al*., 2005) and 1.0% in 206 cats in Colorado, USA (Hill *et al*., 2000). *Salmonella* carrier state in most cats is clinically inapparent and

prevalence of carriers is variable, with isolation rates of *Salmonella* spp. ranged from 0.0% to 14.0% (Ikeda *et al*., 1986).

In a study on the prevalence of enteric zoonotic organisms in cats in Colorado, USA found that shelter cats (18.2%) had higher proportion compared than client-owned cats (10.1%) had enteric organisms as reported by Hill *et al.* (2000), in which 1.9% of shelter cats shed *Salmonella* spp. and non from household cats. Stray carnivores are often exposed to intestinal infection with *Salmonella* species and might remain carriers for long period, so they have great possibilities for shedding these organisms; thus, the stray cats might contribute actively in contamination of environment (Zenad *et al.*, 2014).

According to Immerseel *et al*. (2004), in another study on cats on the risk of transmission of antimicrobial drug-resistant *Salmonella* in Belgium found that grouped-house cats (51.4%) had highest proportion of *Salmonella* isolated followed by diseased cats (8.6%) and healthy house cats (0.36%). As the *Salmonella* organisms commonly inhibit the digestive tract and their associated lymph nodes of infected cats, they excrete *Salmonella* organisms intermittently in their faeces (Gallaway *et al*., 2008; Carter and Quinn, 2000). The age of these animals may also play a role, as young animals are more susceptible to *Salmonella* infection (Immerseel *et al*., 2004). Some researchers considered the apparently healthy cats pose low risk on public health (Wilson, 2004).

Not all cats infected with salmonellosis will shows clinical sign. This is supported by Polpakdee *et al*. (2012) reported in a study of epidemiology and antimicrobial resistance of *Salmonella* spp. isolated from dogs and cats in

Northeastern Thailand, the *Salmonella* isolation from diarrheal (8%) cats was lower than non-diarrheal (10%) cats. In spite of infected cats being asymptomatic they can perpetuate the *Salmonella* species in their bodies, besides an increase dissemination of salmonellae occurs when they are exposed to stress factors or concurrent diseases (Bhaiyat *et al*., 2009) and prolong uses of antimicrobial drugs (Mather *et al*., 2013), therefore carriers actively contribute to contamination of environment. Companion animal facilities may serve as foci of transmission for salmonellae between animals and humans if adequate infection control measures are not followed (Wright *et al*., 2005).

For antibiotic sensitivity test, all three (100%) isolates were resistant to ampicillin, but were susceptible to ciprofloxacin, and one (33%) isolate was susceptible to gentamicin and chloramphenicol. Any misuse of antibiotic administration may cause an infecting organism or even commensals to acquire transferable (plasmid-mediated resistance) resistance, which is of public health significance as such resistance has been demonstrated amongst *Salmonella* isolates from dogs to humans (Greene, 1998). Therapeutic failure could therefore result when the antibiotic is used in infected individuals. In the study on cats in Belgium found that, the *S.* Typhimurium isolated from cats were resistant to single drug such as ampicillin or chloramphenicol, while most isolates from group-housed cats were resistant to ampicillin, chloramphenicol and tetracycline (Immerseel *et al*., 2004), in which the isolated serotype indicate that the cats were infected from the same sources compared with other animals and man. Multidrug-resistant *Salmonella* Typhimurium have been associated with outbreaks of gastrointestinal infections in companion

animals in veterinary clinics and animal shelters (Wright *et al.,* 2005). Increasing antimicrobial resistance among *Enterobacteriaceae* is emerging as a significant public health concern in human medicine (Pitout and Laupland, 2008).

6.0 CONCLUSION

This study suggested that the stray cats are potential carriers of antibiotic resistant *Salmonella* spp. Cats that freely roam outside to scavenge or hunt for food are more exposed to *Salmonella* infection compared with client-owned cats. Free roamer and stray cats are potential candidates for *Salmonella* carriage and might contribute actively to the contamination of environment.

Even though there were none client-owned cats positive for *Salmonella* spp., precautions need to be taken when handling with pets especially household with immunocompromised family members, children as well as aged family members to prevent transmission occur when cats got infected at upon of time after have contact with other infected cats. A large number of *Salmonella* may be shed in faeces; recovered pets may shed *Salmonella* intermittently for months or longer as a recovered carrier. A "carrier state" is one in which the animal has no signs of disease but harbors *Salmonella* and can transmit it to other animals or people. Stray cats pose a potential threat to public health and their faecal materials may play significant roles in the contamination of environment.

7.0 RECOMMENDATION

There are key points of transmission of *Salmonella* spp. and antibiotic resistant organisms from cats to human:

1. People should wash their hand thoroughly with aseptic wash after coming in contact with animals.
2. Restrict access to the pet in sudden (acute) stage of the disease.

3. The sleeping compound of pets should be kept clean and wash daily with detergent to prevent transmission of pathogens especially household with young children.

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9.0 APPENDIX

Client Consent Form

CLIENT / RESPONDENT'S INFORMATION SHEET AND CONSENT

PENERANGAN DAN PERSETUJUAN PELANGGAN / RESPONDEN

Sila baca maklumat berikut dengan teliti. Sekiranya anda mempunyai sebarang pertanyaan, sila kemukakan kepada penyelidik.

Study is conducted by **Department of Veterinary Pathology & Microbiology, Faculty of Veterinary Medicine, UPM**.

*Kajan ini dijalankan oleh **Jabatan Pathologi dan Microbiologi Veterinar, Fakulti Perubatan Veterinar, UPM**.*

1. STUDY TITLE AND PERIOD STUDY WILL BE CONDUCTED / *TAJUK KAJIAN DAN JANGKA MASA KAJIAN DIJALANKAN*

Prevalence of *Salmonella* in stray and owned cats (*Felis catus*). This study will be held from 9th January 2016 until 22nd February 2016.

Kelaziman bacteria Salmonella pada kucing liar dan kucing kepunyaan orang (Felis catus). Kajian ini akan dijalankan pada 9 Januari 2016 sehingga 22 Februari 2016.

2. PURPOSE AND BENEFIT OF STUDY / *TUJUAN DAN MANFAAT KAJIAN*

From this study, if there is evidence of infection or antibiotic residue for Salmonellosis in the stray or owned cats, it may become carrier for which could harm the public health. Using this study, appropriate measure such as management program could be initiated for cats to ensure free Salmonellosis infection in local cats both owned and strays.

Thus this study will be carried out to determine the current status of prevalence of bacteria and anti-microbial sensitivity on cats.

Daripada kajian ini, jika terdapat bukti jangkitan atau sisa antibiotik untuk Salmonella dalam kucing liar atau dimiliki, ia boleh menjadi pembawa yang mana boleh memudaratkan kesihatan awam. Menggunakan kajian ini, langkah yang sesuai seperti program pengurusan boleh dimulakan untuk kucing untuk memastikan tiada jangkitan Salmonella di kucing tempatan yang dimiliki dan sesat.

Oleh itu, kajian ini akan dijalankan untuk menentukan status semasa kelaziman bakteria dan anti-mikrob sensitiviti pada kucing.

3. SAMPLES REQUIRES FROM YOUR PET/ *SAMPEL DIPERLUKAN DARI HAIWAN KESAYANGAN ANDA*

For this study, faecal samples are required. Therefore, rectal swabs will be taken on your pet with the assistance of veterinarian-in-charge.

Untuk kajian ini, sampel najis diperlukan. Oleh itu, calitan rektum akan diambil pada haiwan kesayangan anda dengan bantuan doktor haiwan yang bertugas.

4. WHO SHOULD YOU CONTACT IF YOU HAVE ADDITIONAL QUESTIONS DURING THE COURSE OF THE RESEARCH? / *SIAPA YANG SAYA PERLU HUBUNGI SEKIRANYA SAYA MEMPUNYAI SOALAN TAMBAHAN SEMASA MENGIKUTI PENYELIDIKAN INI?*

Please contact Ms. Nur Farawahidah Mohsin (Tel: 017-7546572) or Prof Dr. Saleha Abd Aziz (Tel: 03-86093458) if you have any questions regarding this research.

Sila hubungi saya, penyelidik kajian, Nur Farawahidah Mohsin (Tel: 017-7546572) atau penyelia kajian, Prof Dr. Saleha Abd Aziz (Tel: 03-86093458) jika anda mempunyai sebarang pertanyaan mengenai kajian ini.

Please contact us the IACUC (03-89471244) if you have any questions regarding this research.

Sila hubungi kami, ahli IACUC (03-89471244) jika anda mempunyai sebarang pertanyaan mengenai kajian ini.

Please sign here if you have read and understood the contents of this page

Sila tandatangan di sini sekiranya anda telah membaca dan memahami kandungan halaman ini

Signature/ *Tandatangan* ..... Signature/ *Tandatangan* .....

(Respondent / Respoden) (Witness / *Saksi*)

Date/ *Tarikh*:..... Name/ *Nama* :.....