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RESEARCH ARTICLE

A Study of Aetiology and Risk Factors of Bacterial Septicaemia of Cats

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A B S T R A C T

Cats have been a popular species of animals as companion animals and their health awareness has become an important issue among both veterinarians and owners. Septicaemia is one of the most important disease in both human and veterinary medicine. This article describes a retrospective study involving septicaemia cases among cats between 2006 and 2016 presented to the Post-mortem Laboratory, Faculty of Veterinary Medicine, Universiti Putra Malaysia. Some of the common microorganisms causing septicaemia in these cats include *Escherichia coli* (41%), *Klebsiella pneumoniae* (14%), *Rhodococcus equi* (13%) *Streptococcus* sp. (11%), *Staphylococcus* sp. (9%), *Pasteurella* sp. (8%), *Salmonella* sp. (2.4%) and *Pseudomonas aeruginosa* (1.6%). Respiratory tract was observed as the most common point of entry of microorganisms leading to septicaemia in cats albeit statistically insignificant (34.7%, P>0.05). These septicaemic cases were highly associated with risk factors such as underlying infection (55.6%), stress (18.5%), malnutrition (18.5%), tumour (5.6%) and traumatic injury (16.9%). Further analysis revealed that underlying viral infection predominates compared to bacterial and parasitic infections. Septicaemia is an important health problem among cats and different agents are associated with different point of entries. The association and the relationship between the aetiological agents, points of entry, and risk factors were further discussed.

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INTRODUCTION

Septicaemia is the presence of microorganism and its toxin in the circulatory system leading to systemic inflammatory response, and eventually death (Remick, 2007). Most of the time septicaemia worsens the case by causing septic shock leading to multiple organs dysfunctions due to profound haemodynamic alterations (De Backer *et al*., 2014). Despite the high expenditure of treatment, sepsis still causes high rate of fatality among humans (Hall *et al*., 2011). Similarly, sepsis is regarded as an important clinical entity with high mortality rate among cats (Babyak and Sharp, 2016). Prior to death, cats suffering from sepsis frequently exhibit clinical signs such as lethargy, pale mucous membrane, abdominal pain, tachypnoea, bradycardia, weak pulses, hypothermia and icterus (Brady *et al*., 2000). Bradycardia and hypothermia are negative prognostic indicator in sepsis cases (Osterbur *et al*., 2014) suggestive of the terminal stage of sepsis. Sepsis is often diagnosed at the later stage due to the acute nature of the disease (Remick, 2007).

Septicaemia is commonly caused by bacterial infection (Osterbur *et al*., 2014) and Gram-negative bacteria predominates among animals (Ramachandran, 2014). Number of companion animals have substantially increased in the modern society attributing to pet welfare awareness (Guardabassi *et al*, 2004). Recently, there are evidences suggestive of growing antimicrobial resistance (AMR) bacteria causing infection in pets (Lloyd, 2007) including *Staphylococcus intermedius*, *E. coli*, methicillin-resistance *Staphylococcus aureus* and other pathogens (Lloyd, 2007). Transmission of these AMR microorganisms among pets, pet owners and veterinary personnel may cause a problem in the future. Determination of causative agents and the risk factors associated with septicaemia is the first step towards addressing this problem. The aim of this study is to identify the common aetiological agents and risk factors associated with septicaemia in cats.

MATERIALS AND METHODS

Data collection: Post-mortem records on cases of cats presented to the Post-mortem Laboratory, Faculty of Veterinary Medicine, Universiti Putra Malaysia (UPM) between 2006 and 2016 were collected. The data collected from each case includes the age, sex, breed, history, necropsy and other laboratory results such as parasite, viral, or bacterial isolation. Cases with no definitive diagnosis, missing, or incomplete reports were not included in the study. Septicaemia was diagnosed based on bacterial isolation with the isolation of the same bacteria from at least 3 samples of internal organs of a carcass showing moderate to severe visceral organs and musculature congestion. The diagnosed cases of septicaemia were analysed for their correlation with sex, age, point of entry, type of microorganism and risk factors. The risk factor of each case of septicaemia was categorised into five categories; underlying infection, tumour, traumatic injury, stress, malnutrition, and miscellaneous (eg. post-surgical complication and multiple organ dysfunction syndrome).

Age: The age of the cats were categorised as previously described by Rathiymaler *et al*. (2017). Briefly, this consist of three groups; pediatrics (≤6 months old), adult (7 months – 10 years), and senior (>10 years). Ratio between number of septicaemia cases and the total number of cases sent for post-mortem laboratory according to the age group was compared for a fair comparison.

Determination of point of entry: Collection of data pertaining to the points of entry was done as reported by resident pathologists. The points of entry were classified based on the body systems includes respiratory, gastrointestinal (GIT), urinary and integumentary systems. The one identified as others may include reproductive, musculoskeletal system or which has unknown point of entry. As the cases of reproductive and musculoskeletal systems have less than 10 cases from the overall septicaemia cases, thus they are classified under others.

Aetiological agents: The bacteriology report obtained for each case determines the causative microorganisms involved in these septicaemia cases. In cases with multiple organisms' isolation, the main causative agent of septicaemia was identified based on the isolation of the same organism from all organs submitted for bacteriological isolation and identification. All bacterial isolations and identification using biochemical testing were made according to methods previously described (Carter and Cole, 2012)

Data analysis: Statistical Packages for the Social Sciences (SPSS) version 22 was used to analyse the data. Frequencies of each categorical group according to the sex, breed and age of the animals were determined. Non-parametric test was used for data analysis as the data were categorical and non-continuous. All tests were done at 95% confidence intervals level. Subsequently, the point of entry and the most common bacteria isolated as well as the association between variables was determined using Pearson's chi-squared test. Excel version 2007 was used to identify the relation between age group to point of entry and age group to risk factors and Pearson's chi- squared test was used to identify the significance between these variables.

RESULTS

Prevalence: A total of 431 cases of cats were submitted for post-mortem examination between the year 2006 and 2016 and 128 (29.7%) of these cases were diagnosed as septicaemia. Four cases (3.1%) were excluded due to inconclusive diagnosis and missing reports, resulting in only the remaining 124 cases were subjected to data analysis. It was found that 72 (58.1%) cases involved male cats and the other 52 (41.9%) cases involved female cats but without significant difference (P=0.811). Similarly, no significant difference (P=0.45) was noted on comparison between different breed of cats diagnosed with septicaemia (Table 1).

Age of animal: Out of 124 cases, 52 (53%) cases involved the paediatrics, 68 (26%) cases involving the adult and 4 cases (6%) involved the senior cats (Table 2). There is highly significant difference (P=0.01) between the paediatrics groups compared to other two age groups among cats that are susceptible to septicaemia.

Table 1: Total number cases for different breed of cats with septicaemia between the year 2006 and 2016.

Breed	Total number of cases of septicaemia	Total number cases of post mortem	Percentage (%)
Domestic Short Hair	85	234	36.3
Persian	18	94	19.2
Maine Coon	7	45	15.6
Bengal	4	21	19.0
Others	10	37	27.0
Total	124	431	28.8

Table 2: Total number cases for each age group of cats with septicaemia between the year 2006 and 2016.

Age group	Total number of cases of septicaemia	Total number cases of post mortem	Percentage (%)
Paediatrics	52 (42%)	98	53.1*
Adult	68 (55%)	264	25.8
Senior	4 (3%)	69	5.8
Total	124 (100%)	431	28.7

Chi-square test, X^2 (2, N=124) = 117.1, P=0.01, * indicates significant difference between the paediatrics groups and the other two age groups among dog that are susceptible to septicaemia.

Point of entry: Analysis of data revealed that 43 (34.7%) cases of septicaemia originates from respiratory tract, followed by 39 (31.5%) cases from GIT, 18 (14.5%) cases from urinary tract and 13 (10.5%) cases from the integument while the remaining cases were from other routes. No significant difference (P=0.15) was noted between these points of entry.

Bacterial isolation: Analysis of bacteriology report revealed that 34 (27.4%) cases resulted in isolation of pure culture of bacteria, 39 (31.5%) cases were had isolation of mixture of two species of bacteria, and 51 (41.1%) cases yielded isolation of three or more species of bacteria. *E. coli* was significantly (P<0.05) more frequently isolated with 51 (41.1%) cases, followed by *K. pneumoniae* (17 cases, 13.7%), *R. equi* (16 cases, 12.9%), *Streptococcus* sp. (14 cases, 11.3%), *Staphylococcus* sp (11 cases, 8.9%), *Pasteurella* sp. (10 cases, 8.1%), *Salmonella* sp. (3 cases, 2.4%), and *Pseudomonas aeruginosa* (2 cases, 1.6%), X^2 (7, N=124)=33.9, P=0.023.

It was found that *Streptococcus* sp. (P=0.034) and *Staphylococcus* sp. (P=0.012) significantly more commonly isolated in mixed-agents sepsis (11 cases, 78.6%, and 9 cases, 81.8 % respectively) compared to single-agent sepsis (3 cases, 21.4%, and 2 cases, 18.2% respectively).

Risk factors: The most common risk factor in causing sepsis among cats reported was underlying infection involving 69 cases (55.6%). Underlying infection was found to be significantly (P=0.041) important compared to stress of 23 cases (18.5%), malnutrition of 23 cases (18.5%), traumatic injury of 21 cases (16.9%), tumour of 7 cases (5.6%) and miscellaneous risk factors of 16 cases (12.9%) [Chi-square test, X^2 (5, N=124)=17.88]. The underlying infection was classified according bacterial (55.1%), viral (43.5%) and parasitic (1.4%) infection. However, no significant difference was observed (P 0.77) [Chi-square test, X^2 (2, N=69)=11.6]. Other common risk factor was stress. Stress is divided into multi-pet household (43.5%), transportation stress (21.7%), boarding stress (8.7%) and secondary stress (26.1%). The secondary stress in this study includes poor nutrition and heat stress.

Association between the points of entry and the types of bacteria isolated: Septicaemia originated from respiratory tract was found to have significantly (p <0.05) more *Pasteurella* spp. and *R. equi* isolations compared to other point of entry. All 16 cases of septicaemia with *R. equi* isolation [Chi-square test, X^2 (4, N=16)=28.05] and 10 cases of *Pasteurella* spp. isolation [Chi-square test, X^2 (4, N=10)=41.33] causing sepsis in cats were originating from respiratory tract. *E. coli*, *K. pneumoniae*, *Staphylococcus* sp. and *Streptococcus* sp. did not show significant correlation (P>0.05) with the points of entry.

Further analysis revealed significant difference between the respiratory and GIT [Fisher's Exact test X^2 (1, N=82)=17.4, P=0.003], between respiratory and urinary tract [X^2 (1, N=61)=71.3, P=0.006] and between respiratory and integumentary tract in successful isolation of *Pasteurella* spp. [X^2 (1, N=56)=58.1, P=0.001]. There was no significant difference between the GIT and integumentary tracts [X^2 (1, N=52)=0.77, P=0.81], between GIT and urinary tract [X^2 (1, N=57)=11.34, P=0.67] and between urinary and integumentary tract [X^2 (1, N=31)=0.64, P=1.26] in the isolation of *Pasteurella* spp. (Table 3).

As for *R. equi* isolation, significant difference was observed in comparison between respiratory and gastrointestinal tracts [Fisher's Exact test X^2 (1, N=82)=45,11, P=0.017], between respiratory and urinary tract [X^2 (1, N=61)=88.6, P=0.003], and between respiratory and integumentary tract in the isolation of *Rhodococcus equi*. [X^2 (1, N=56)=71.9, P=0.012]. There were no significant differences between the GIT and integumentary tracts [X^2 (1, N=52)=17.89, P=0.55], between GIT and urinary tract [X^2 (1, N=57)=29.2 P=1.74] and between urinary and integumentary tract [X^2 (1, N=31)=42.7, P=0.65] in the isolation of *R. equi*.

Out of 92 cases of septicaemia with *E. coli* isolation, there was no significant (P=0.75) difference compared among the different point of entries; GIT (38 cases, 41.3%), respiratory tract (36 cases, 39.1%), urinary tract (12 cases, 13%) and integumentary tracts (2 cases, 2 %) (Table 3). However, while compared between cases between the point of entry with and without *E. coli* isolation; there was significant difference between the respiratory and integumentary tract [Fisher's Exact test X^2 (1, N=56)=91.4 P=0.004], between GIT and integumentary tract [X^2 (1, N=52)=62.1 P=0.009] and between urinary and integumentary tract [X^2 (1, N=31)=74.5 P=0.048]. There was no significant difference between the respiratory tract and GIT [Chi square X^2 (1, N=82)=69.3, P=1.88], between respiratory and urinary tract [Fisher's Exact test X^2 (1, N=61)=33.5 P=0.87] and between GIT and urinary tract [X^2 (1, N=57)=49.1 P=0.97] in *E. coli* isolation.

Table 3: Total number of each point of entry associated with different types of bacteria in septicaemia cases among cats between the year 2006 and 2016

	E. coli	*K. pneumoniae*	*R. equi*	*Staphylococcus* sp	*Streptococcus* sp	*Pasteurella* sp	Total
GIT	38	12	2	2	5	0	59
Respiratory	36	14	16*	3	4	10*	83
Urinary	12	13	3	1	2	2	33
Integument	2	13	2	1	2	1	21
Others	8	3	0	0	1	1	13
Total	96	55	23	7	14	14	209

R. equi; Chi-square test X^2(4, N=16) = 28.05, p = 0.032. *Pasteurella* sp.; Chi-square test X^2(4, N=18) = 41.33, p = 0.041;* indicates significant difference at P<0.05.

Table 4: Total number of cases and percentage of points of entry associated with their respective risk factors among the between the year 2006 and 2016

	Underlying infection	Malnutrition	Trauma	Tumour	Stress	Miscellaneous	Total
GIT	23 (33.3%)	2 (20%)*	5 (23.8%)	3(28.6%)	10 (43.5%)	5 (7%)	48 (100%)
Respiratory	23 (33.3%)	5 (50%)	5 (23.8%)	2 (28.6%)	10 (43.5%)	5 (16.1%)	50 (100%)
Urinary	13 (18.8%)	1 (10%)	0 (0%)	1 (14.3%)	0 (0%)	0 (14.3%)	15 (100%)
Integument	4 (5.8%)	1 (10%)	9 (42.8%)*	1 14.30%)	2 (8.7%)	1 (12.5%)	18 (100%)
Others	6 (8.7%)	1(10%)	2 (9.5%)	0 (0%)	1 (4.3%)	2 (13.3%)	12 (100%)

Malnutrition; Chi-square test X^2(4, N=23) = 81.09, p = 0.011. Traumatic injury; Chi-square test X^2(4, N=21) = 40.99, p = 0.004; * indicates significant difference at P<0.05.

Table 5: Total number of cases with percentage of age groups according to risk factors of septicaemia among cats between the year 2006 and 2016

	Pediatrics	Adult	Geriatrics	Total
Underlying infection	33 (63.5%)*	34 (50%)	2 (50%)	69
Traumatic injury	8 (15.4%)	13 (19.1 %)	0 (0%)	21
Malnutrition	6 (11.5%)	4 (6%)	0 (0%)	10
Tumor	3 (5.8%)	3 (4.4%)	1 (25%)	7
Stress	21 (40.4%)*	2 (3%)	1 (25%)	23

Underlying infection; Chi-square test $X^2(2, N=69) = 117.08$, $p = 0.001$. Stress; Chi-square test $X^2(2, N=23) = 67.10$, $p = 0.018$;* indicates significant difference at $p<0.05$ for paediatric group compared to the other two age group.

Table 6: Different aetilogical agent causing septicaemia according to each age groups among cats between the year 2006 and 2016

	Paediatrics	Adult	Geriatrics	Total
Bacterial	4 (9.3%)	32 (81.4%)	2 (100%)	38 (100%)
Viral	28 (87%)*	2 (13%)	0 (0%)	30 (100%)
Parasite	1 (100%)	0 (0%)	0 (0%)	1 (100%)
Total	33	34	2	69 (100%)

Chi-square test X^2 (2, N=30) = 95.47, P=0.001, * indicates that its significant compared to the other two group compared for that particular aetiological agent.

Association between the point of entry and risk factors: The association between the point of entry and risk factors are tabulated in Table 4. Traumatic injuries shows a significant (P<0.05) association with integumentary system as the point of entry for septicaemia among these cats. There were no significant association traumatic injury to GIT [Chi-square test X^2 (1, N=52) =56.3, P=0.007], urinary system [X^2 (1, N=31)=79.2, P=0.005], and respiratory [X^2 (1, N=56)=34.6, P=0.002] in causing sepsis. On the other hand, malnutrition shows a significant (P<0.05) association with the GIT as the point of entry for these septicaemic cats compared to respiratory system [Chi-square test X^2 (1, N=82)=52.89, P=0.002], urinary system [Chi-square test X^2 (1, N=57)=66.32, P=0.012] and integumentary system [Chi-square test X^2 (1, N=52)=91.76, P=0.038]. Other risk factors did not show significant associated with the point of entry.

Association between age group and risk factors: The paediatrics group showed the significant (P=0.039) correlation with viral infections than the adult and senior groups. The risk factor of viral (85%) infection significantly (P=0.026) leads to sepsis among the paediatrics compared to bacterial (12%) and parasitic (3%) infections (Table 5).

DISCUSSION

This study suggests that there was no influence of breed and sex of the animal to the occurrence of septicaemia. There is equal chance between female and male cats in acquiring septicaemia. However, paediatric cats are more susceptible to septicaemia compared to the adult and senior cats. Paediatric cats are known to be immunologically more susceptible towards clinical septicaemic salmonellosis and bacterial infection compared to healthy adult cats (Stiver *et al*., 2003). Besides, this study found that paediatrics are more susceptible to viral infections, which act as an important risk factor to septicaemia. Diseases of the upper respiratory tract such as by feline herpesvirus and feline calicivirus have been observed to have high prevalence among paediatric cats (Binns *et al*., 2000). Previous study suggested high susceptibility of paediatric cats towards various conditions such as pyothorax (Waddell *et al*., 2002; Brady *et al*., 2000) and bacterial ileocolitis (De Cock *et al*., 2004) due to under-developed immunity and challenges in new environment (Waddell *et al*., 2002). Thus, this explains why paediatrics are more susceptible in acquiring septicaemia compared to adult and senior cats.

This study reveals that underlying infection from any body system significantly contribute to septicaemia compared to other risk factors. This is possibly due to the presence bacteria or pathogens from the underlying infections facilitate bacterial proliferation leading to septicaemia. As concluded by previous study, the most common risk factors of sepsis in cats is associated with underlying infection such as pyothorax, septic peritonitis, bacteraemia secondary to gastrointestinal disease, pneumonia, endocarditis, osteomyelitis, pyometra, bite wound and pyelonephritis (Brady *et al*., 2000). Logically we need the presence of the causative agent to start and spread the infection (Remick, 2007), thus presence of underlying infection causes immunological disturbance which favours infection to take place.

As for the microorganism causing sepsis in this study, the isolation of *E. coli* was significantly high compared to other species of bacteria. We found out that the gastrointestinal, respiratory and urinary tracts have significantly higher isolation of *E. coli* compared to the integumentary tract. A similar trend has been observed from previous study on infection and microbiology among cats, where *E. coli* has the highest isolation rate from feline lower urinary tract (Litster *et al*., 2007), and GIT (Costello *et al*., 2004). Besides, in another study, even though *E. coli* is a pathogen of non-oropharyngeal origin but it was successfully isolated from feline pyothorax along with *Klebsiella* sp. and *Pasteurella* sp. (Barrs *et al*., 2005). This suggest that *E. coli* can been easily isolated from these routes thus sepsis involving *E. coli* from these tracts in our study is possible. This is similar in septicaemia cases of dog from a previous study reveals that *E. coli* has been significantly isolated compared to other type of bacteria (Rathiymaler *et al*., 2017). There were also studies reported in foals and calves, where *E. coli* predominates in causing neonatal sepsis (McKenzie and Furr, 2001). It can be said that *E. coli* is well known in causing infection and sepsis in many species of animals and cat is not an exception according to this study.

The most common point of entry of septicaemia in cats is the respiratory tract. This is explained by the fact that cats are very susceptible to lung infection and respiratory distress (Stillion and Letendre, 2015). In our study, *Pasteurella* spp. was predominately isolated in respiratory tract but not in GIT, urinary and integumentary tracts. Possibly, most cats carry these organisms in their respiratory tract as normal flora (Kimura *et al*., 2004). In other species of animals, these bacteria may proliferate when the host underwent stressful episodes, and eventually causing septicaemic disease (Fitri *et al*., 2017; Annas *et al*., 2014). The theory of carrier status is being supported by reports of *P. multocida* septicaemia among human that has close contact with cats. This shows that cats have *Pasteurella* sp. in their nasopharyngeal area and can transmit through secretion droplet through close contact (Orsini *et al*., 2013). The importance of *Pasteurella* sp. in association with the respiratory infections among cats was previously reported (Stillion and Letendre, 2015).

The other species of bacteria frequently isolated from cases of septicaemia with respiratory tract origin is *R. equi*. Sporadic cases of *R. equi* infections in cats is known to be associated with pneumonia (Farias *et al*., 2007). *R. equi* may gain entry into the host following exposure to contaminated manure or soil, or direct contact with infected animals either via aerosol or cutaneous route (Takai *et al*., 2003). *R equi* ability to be disseminated aerosolly leads to this organism being commonly causing septicaemia via the respiratory route (Farias *et al*., 2007). These justify the reason *R. equi* predominate in respiratory tract but not in GIT, urinary and integumentary tracts according to our study. Basically this explains *R. equi* colonize the respiratory tract, particularly the lungs as site of multiplication.

Based on our study, the significant correlation between traumatic injury (risk factor) and integument (route of entry) was observed understandably due to the fact that the skin functions as the first line barrier to outside environment. Breaching of the skin barrier allows pathogens to enter the host immune system to cause septicaemia (Remick, 2007). Pathogens take chances every time there is imbalance of the immune system, thus damage to the skin causing any pathogen to invade the host system. Mostly, traumatic injuries involve physical damage and the first line that will be affected will be the integument compared to other organs and system. Hence, skin injuries in cat significantly related to septicaemia compared to other point of entry.

Conclusions: Septicaemia is a crucial health issue among cats as it causes high mortality rate and major public health related to transmission of antimicrobial resistance. *E.coli* is highly isolated from these cases and can be originated from the GIT, respiratory and urinary tracts. *Pasteurella* sp and *R. equi* is significantly isolated from respiratory tract which is the common point of entry among these cats. The paediatrics has high susceptibility to viral infection leading to higher susceptibility to sepsis, thus veterinary clinics should encourage pet owners to vaccinate and perform health check-ups for their kittens.

Authors contribution: RM involved in data collection, statistical analysis, and drafting of manuscript. AS and ZM involved in supervision, study conception and design, interpretation and manuscript revision. JFFA and ZZ involved in supervision and manuscript revision.

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