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Prevalence of Bacteriological Isolates Recovered from Faecal Sample of Domestic Cats

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A B S T R A C T

The study was performed for prevalence of bacteriological isolates recovered from faecal samples of domestic cats and their antibiotic sensitivity pattern. Total thirty rectal swabs were collected from Baroda for bacteriological isolation. The overall prevalence of bacteria was 93.33% (28). The prevalence of *E. coli*, *Streptococcus* spp., *Staphylococcus* spp., gram positive bacilli, gram negative coccobacilli and gram positive coccobacilli were 36.67% (11), 23.33% (7), 10.00% (3), 10.00% (3), 10.00% (3) and 03.33% (1) respectively. *In-vitro* antibiotic sensitivity pattern of all the bacterial isolates recovered from faecal samples of domestic cats revealed that the isolates were more sensitive to Gentamicin (71.43%) followed by Co-trimoxazole (57.14%), Colistin (33.33%), Enrofloxacin and Amoxyclav (28.57% each), Tetracycline (19.05%) and Ampicillin (14.29%).

Introduction

The cat (*Felis catus*) is also known as domestic cat or house cat to distinguish it from the felines and felids.

It is a small furry domesticated carnivorous mammal that is valued by humans for its companionship household pets. Cats have been associated with humans for at least 9,500 years, and are currently the most popular pet in the world (Driscoll *et al.*, 2009). Owing to their close association with humans, cats are now found almost everywhere in the world. Cats are the most favorite pets after dogs.

The intestinal microbiota is the collection of the living microorganisms (bacteria, fungi, protozoa, and viruses) inhabiting the gastrointestinal (GI) tract. Novel bacterial identification approaches have revealed that the gastrointestinal microbiota of dogs and cats is, similarly to humans, a highly complex ecosystem, comprising at least several hundred different bacterial phylotypes (Suchodolski *et al.*, 2009). It has been suggested that the intestine of mammals is home to a total of 1010-1014 microbial cells, which is approximately 10 times more than the number of host cells. This complex microbial ecosystem and its interplay with

eukaryotic host cells have a significant impact on health and disease of cats.

Based on traditional bacterial culture, the small intestine of dogs and cats harbors generally low bacterial counts, ranging between 102 to 105 cfu/g of small intestinal content; Cats appear to have higher counts of anaerobic bacteria compared to dogs in the proximal small intestine (Johnston *et al.,* 1993). The total bacterial count in the colon ranges between approximately 109 and 1011 cfu/g and the most abundant cultivable groups are *Bacteroides, Clostridium, Lactobacillus, Bifidobacterium*, and *Enterobacteriaceae* (Mentula *et al.,* 2005).

On average, 10 different bacterial phyla have been identified in the feline gut, with *Firmicutes, Bacteroidetes, Proteobacteria, Fusobacteria,* and *Actinobacteria* making up the vast majority of all gut microbes (Desai *et al.,* 2009). Minor abundant members are the phyla *Tenericutes, Verrucomicrobia, Cyanobacteria,* and *Chloroflexi*. The *Firmicutes* contain various sequences affiliated with *Clostridium* cluster IV and *Clostridium* cluster XIVa and these are together with *Bacteroides* or *Prevotella* the predominant bacterial groups in fecal samples (Handl *et al.,* 2011). *Helicobacter* are the predominant group in the stomach (> 90% of sequencing reads) (Wagner, 2008), while the duodenum is home to *Enterobacteriaceae, Clostridiales, Bacteroidales,* and *Lactobacillales.*

The gastrointestinal microbiota has a strong impact on the health of cats and the microbiome can be altered in GI disease (Bell *et al.,* 2008). The intestinal microbiota plays a crucial role in the development of the host immune system, protection against pathogens, toxins and mutagens and utilization of excess nutrients or nutrients that are unavailable to the host (Inness *et al.,* 2007). Alterations of the normal gut microbiota balance due to inherent, environmental or immunological factors can be involved in the pathogenesis of intestinal inflammatory diseases.

Materials and Methods

Collection

During survey work total 30 rectal swabs were aseptically collected from domestic cats with help of sterile swab.

Isolation

The rectal swab was inoculated on MacConkey agar and blood agar for primary isolation of bacteria and later on sub culture on MacConkey agar and Eosine Methylene Blue (EMB) agar for isolation and identification.

The inoculated plates were examined for morphological, characteristics and growth of bacterial colonies after 24-48h incubation period. The isolates were then identified on the basis of colony characteristics, staining characteristics (after staining with Gram's stain) microscopic morphology, lactose fermenting ability on MacConkey agar and greenish metallic sheen on EMB agar.

The Gram-positive cocci in chains were identified as *Streptococcus* spp. and in cluster were identified as *Staphylococcus* spp. on the basis of morphological characteristics. The gram negative coccobacilli isolates recovered on MacConkey agar with lactose fermenting pink colour colony on the culture plates. The greenish metallic sheen colonies were identified as *E. coli*on EMB agar plates.

Antibiotic sensitivity test

All the isolates confirmed by primary test were subjected for antimicrobial sensitivity

test. The antimicrobial sensitivity test was carried out by disc diffusion technique of Bauer *et al.,* (1966) using seven antimicrobials. Mueller-Hinton agar plates were prepared, and the samples were inoculated. Antibiotic discs were placed at specific distance, and the plates were incubated at 37°C for 24 h. The results were read by measuring the zone of inhibition produced by various antibiotic discs and compared to the standards. The antibiotic discs used were ampicillin (AMP; 10 mcg), gentamicin (GEN; 10 mcg), enrofloxacin (EX; 10 mcg), co-trimoxazole (COT; 25 mcg), colistin (methane sulphonate) (CL; 10 mcg), amoxyclav (amoxycillin-clavulanic acid) (AMC; 30 mcg), and tetracycline (30 mcg).

Results and Discussion

Bacteriological isolates recovered from faecal samples of domestic cats

In the present investigation, bacteriological culture examination of rectal swabs from 30 cats, resulted the recovery of a total 28 bacterial isolates either in pure culture and/or as part of mixed infection. The bacteriological prevalence from faecal samples of domestic cats is presented in Table 1. The overall prevalence of bacteria was 93.33% (28/30). Of these, monomicrobic isolates were recovered from76.67% (23/30) cats whereas more than one bacteria were found in case of 16.67% (5/30) cats. *E. coli* was found as a major isolate from faeces of cats. Total 11 cats were infected with *E. coli* infection. Out of total (n=11) positive samples (n=5) samples were mixed with *Streptococcus* spp. The prevalence of *E. coli* was 36.67% (11/30). *Streptococcus* spp. was recovered from (n=7) samples of domestic cats, out of them (n=5) samples were recovered as mixed isolates along with *E. coli*. The prevalence of *Streptococcus* spp. was 23.33% (7/30).Out of 30 samples *Staphylococcus* spp. was recovered from (n=3) faecal samples with the prevalence of 10.00% in domestic cats. Gram positive bacilli was also recovered from (n=3) feacal samples with the prevalence of 10.00%.Gram negative coccobacilli was isolated from (n=3) faecal samples with the prevalence of 10.00%.Gram positive coccobacilli was also recovered from (n=1) faecal sample and the prevalence was 03.33% in domestic cats.

In-vitro antibiotic sensitivity test

Total 28 bacterial isolates recovered from faecal samples of domestic cats. Out of 28 isolates, 21isolates were subjected for *in-vitro* antibiotic sensitivity test. *In-vitro* antibiotic sensitivity pattern of all the 21 bacterial isolates recovered from faecal samples of domestic cats were shown in Table 2 and Figure 1. *In-vitro* antibiotic sensitivity pattern of all the bacterial isolates recovered from faecal samples of domestic cats revealed that the isolates were more sensitive to gentamicin (71.43%) followed by co-trimoxazole (57.14%), colistin (33.33%), enrofloxacin and amoxyclav (28.57% each), tetracycline (19.05%) and ampicillin (14.29%).*E. coli* isolates were highly sensitive to colistin (100%) followed by gentamicin (66.67%), co-trimoxazole (50.00%) and the least to amoxyclav (16.67%). ampicillin, enrofloxacin and tetracycline drugs were reported as highly resistant drugs (100%) against *E. coli* isolates. *Streptococcus* spp. was highly susceptible to enrofloxacin (100%), followed by tetracycline (60.00%), enrofloxacin, co-trimoxazole, and amoxyclav (40.00% per cent each) and ampicillin (20.00%). Colistin drug was reported as highly resistant drug (100%) against *Streptococcus* spp. *Staphylococcus* spp. isolates were higher susceptible to gentamicin (66.67%), followed by co-trimoxazole, enrofloxacin and amoxyclav drugs (33.33% each). Colistin, ampicillin and tetracycline reported as highly resistant drugs (100%) against *Staphylococcus* spp. isolates.

Gram positive bacilli isolates were highly sensitive to gentamicin (100%) followed by co-trimoxazole andamoxyclav (66.67% each) and tetracycline (33.33%). Ampicillin, enrofloxacin, and colistin drugs were reported as highly resistant drugs (100%) against Gram positive bacilli isolates. Gram negative coccobacilli isolates werehighly susceptible to co-trimoxazole (100%), followed by ampicillin and enrofloxacin (66.67% each), gentamicin and colistin (33.33% each). Amoxyclav and tetracycline were highly resistant drugs (100%) against Gram negative coccobacilli. In present study Gram positive coccobacilli isolates were highly sensitive to gentamicin, enrofloxacin and co-trimoxazole (100% each). Ampicillin, colistin, amoxyclav and tetracycline drugs were reported as highly resistant drugs (100%) against Gram positive coccobacilli isolates.

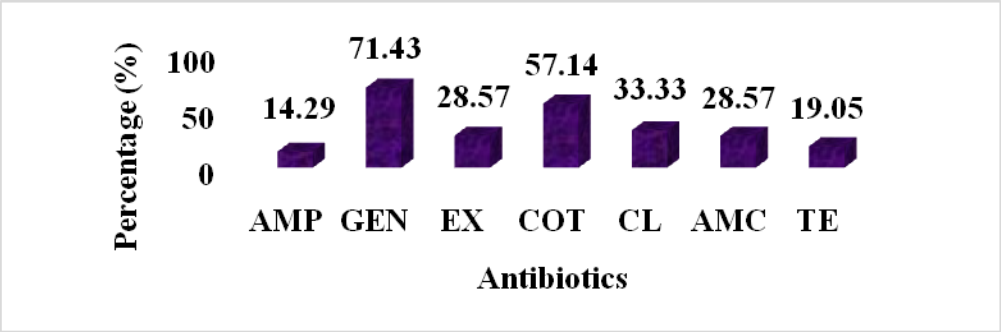
Table.1 Bacteriological prevalence from faecal samples of domestic cats

Name of Bacteria	Total no. of animals screened and bacteriological prevalence rate
N = 30	
A. *Streptococcus* spp.	7 (23.33%)
B. *Staphylococcus* spp.	3 (10.00%)
C. *E.coli*	11 (36.67%)
D. Gram positive bacilli	3 (10.00%)
E. Gram negative coccobacilli	3 (10.00%)
F. Gram positive coccobacilli	1 (3.33%)
Total	**28 (93.33%)**

Table.2 Summary of bacterial isolates recovered from faecal samples of domestic cats and their Antibiogram pattern

Sr. No.	Organism	No. of Isolates	SENSITIVITY (%)						
			AMP	GEN	EX	COT	CL	AMC	TE
1	***E. coli***	6	00.00%	66.67%	00.00%	50.00%	100%	16.67%	00.00%
2	***Streptococcus* spp.**	5	20.00%	100%	40.00%	40.00%	00.00%	40.00	60.00%
3	***Staphylococcus* spp**	3	00.00%	66.67%	33.33%	33.33%	00.00%	33.33%	00.00%
4	**Gram positive bacilli**	3	00.00%	100%	00.00%	66.67%	00.00%	66.67%	33.33%
5	**Gram negative coccobacilli**	3	66.67%	33.33%	66.67%	100%	33.33%	00.00%	00.00%
6	**Gram positive coccobacilli**	1	00.00%	100%	100%	100%	00.00%	00.00%	00.00%

Fig.1 *In vitro* antibiotic pattern to all bacterial isolates recovered from faecal samples of domestic cats



In present study the prevalence of *E. coli* was 36.67% (11). Apart from these communications, Costa *et al.,* (2008) have also reported that two *Escherichia coli* isolates per sample were recovered (66 of cats). Gumus *et al.,* (2017) isolated *E. coli* from rectal swabs of 192 healthy cats, and 82 *E. coli* were isolated (n=82). All samples were incubated for 18 h at 37°C in tryptic soy broth (TSB) and subcultured on MacConkey agar. In present study the prevalence of *Streptococcus* spp. was 23.33% (7). Lysková *et al.,* (2007) also recovered *Streptococcus* spp. from 4 samples of 34 faecal samples. The prevalence of *Staphylococcus* spp. in this study was 10.00% (3). Bierowiec *et al.,* (2016) reported the prevalence of *Staphylococcus aureus* from faecal samples of cats were 17.5% (7). In present study the prevalence of gram positive bacilli was 10.00% (3). Andrzejewska *et al.,* (2013) isolated 7 *Campylobacter* spp. from 71 cats from Bydgoszcz region, and prevalence was 9.86%. Polzler *et al.,* (2018) also recovered *Campylobacter* from 344 cats in Styria, Austria and prevalence of *Campylobacter* spp. was 22 (6.4%).The differences in the occurrence of bacteria might be due to the differences in epidemiological conditions between countries. The antibiotic susceptibility patterns of all isolates found in our study are generally in agreement with the findings of other authors Abdallah (2005) reported that *E.coli* isolates were 47% resistant to tobramycin, gentamycin, penicillin, tetramycin, cefoperazone, erythromycin, chloramphenicol, sulfa, trimethoprim, doxycycline, rifampicin, streptomycin, ofloxacin, cephradine, ceftriaxane, cefotaxime, clindamycin, ampicillin, amoxicillin, nitrofurantoin, norfloxacin, and carbenicillin. Murphy *et al.,* (2009) reported the prevalence of antimicrobial resistance in *E. coli* was as follows: streptomycin (2%), ampicillin (4%), cephalothrin (1%), and tetracycline (2%). Kanagarajah *et al.,* (2017) reported that feline MRSA isolates resistance were as follows, methicillin (100%), ceftazidime (100%), enrofloxacin (92.31%), oxacillin (84.62%) and vancomycin (0%), 84.62% of feline MRSA isolates indicated resistance to four out of five antibiotics tested. Lazou *et al.,* (2017) reported that all *Campylobacter* isolates exhibited susceptibility to erythromycin, gentamicin and streptomycin. Contrariwise, 4.5% of feline isolates were resistant to quinolones, quinolones along with tetracycline and tetracycline alone, respectively. Rodrigues *et al.,* (2015) reported that more than 50% of the *Campylobacter* spp. resistant to ceftiofur, sulphazotrim, norfloxacin and tetracycline. In this study, all bacterial isolates were more resistance to ampicillin and tetracycline. However, due to increasing resistance of bacterial isolates to

antimicrobial agents, there is need to emphasize the importance of susceptibility testing in order to establish correct therapeutic protocol.

In conclusions ampicillin was found to be more resistant on faecal sample so in such cases avoid the use of ampicillin in intestinal infection. Gentamicin was sensitive on faecal sample culture followed by co-trimaxazole, colistin, enrofloxacin, amoxyclav and tetracycline.

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