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Trends in development of antibiotic resistance in *E. coli, Staphylococcus aureus and Pseudomonas aeruginosa* from dogs of Chhattisgarh

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Abstract
The present investigation was carried out to study changing pattern of antibiotic resistance in bacterial isolates namely *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *E. coli* obtained from dogs in Chhattisgarh. Representative samples were collected from dogs presented with clinical conditions *viz.* urinary tract infection, otitis externa and chronic pyoderma during two different periods *i.e.* 2013-14 and 2016-17. Samples were subjected to bacterial isolation and antibiotic disc diffusion test as per standard protocol. Predominant isolates in urinary tract infection and pyoderma were *E. coli* and *Staphylococcus aureus* during both the periods of study. However in cases of otitis externa, changing trend was reported; *Staphylococcus aureus* being predominant isolate in 2013-14 and *Pseudomonas aeruginosa* during 2016-17. Multidrug resiastant *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E. coli* were reported during both the phase of present study. Isolates showed relatively increased sensitivity towards imipenam. There was generous increase in resistance of *Staphylococcus aureus* for amoxyclav, amoxicillin, ceftazidime, ceftriaxone and gentamicin during 2016-17 as compared to 2013-14 isolates whereas changing pattern of increased sensitivity of *Staphylococcus aureus* isolates to ciprofloxacin and chloramphenicol was reported. *E. coli* showed increased resistance for ciprofloxacin, amoxyclav, amoxicillin and ceftriaxone during 2016-17 as compared with 2013-14. There was a harsh increase in antibiotic resistance among *Pseudomonas aeruginosa* for antibiotics namely ciprofloxacin, amoxyclav, imipenam, azithromycin, ampicillin and ceftriaxone. Increased incidence of antibiotic resistance observed in the present study poses a serious threat to livestock and also has public health significanc.

Keywords: Antimicrobial drug resistance, *Staphylococcus aureus, E. coli, Pseudomonas aeruginosa*, dogs

Introduction
Urinary tract infections (UTIs), otitis externa and pyoderma are the common problems in present scenario to the dog owners and pet practitioners (Roy *et al.*, 2018; Seckerdieck *et al.* 2018 and Passmore *et al.*, 2007) [25, 28, 21]. Recurrence of these conditions after cessation of antibiotic therapy, usually within a month highlighted the existence of multidrug resistant isolates in canine veterinary practice.

Though, a variety of bacterial pathogens are regarded as the cause of UTIs, otitis externa and pyoderma in dogs; *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *E. coli* have significant contribution in recurrence of clinical conditions due to intrinsic antibiotic resistance in *Pseudomonas aeruginosa* (Mansour *et al.*, 2013) [17] and emergence of multidrug resistance in *Staphylococcus aureus (*Benito *et al.*, 2016) [4] and *E. coli* (Wong *et al.*, 2015) [34], which underscore the need for rationale antibiotic therapy. Antimicrobial resistance is not only threat to animal medicine but also have great public health concern because direct contact between animals and human can lead to interspecies transmission of pathogenic bacteria including strains that demonstrate antimicrobial resistance (Llyod, 2007) [14].

The present investigation was therefore designed in retrospective way to investigate and compare antibiotic resistance pattern of bacterial isolates namely *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *E. coli* obtained from dogs during two different periods *i.e.* 2013-14 and 2016-17 so as to put forth the observation for rational therapy in Chhattisgarh.

Materials and Methods
Location of Study
Present investigation was conducted at Department of Veterinary Microbiology, Veterinary College, Anjora, Durg (Chhattisgarh).

Collection of samples
Clinical cases including urinary tract infection (UTIs), chronic recurrent pyoderma and otitis externa in dogs were attended and clinically investigated as and when reported from different regions of Chhattisgarh at TVCC, Veterinary College, Anjora, Durg and district veterinary hospitals in Chhattisgarh during two different course of study (2013-14 and 2016-17). Representative samples including urine (25 and 30), ear secretion (30) and pus (25 and 20) were collected aseptically for bacteriological examination during 2013-14 and 2016-17, respectively.

Sample processing
Urine samples were centrifuged at 1500 rpm for 15 minutes. The sediments were diluted with sterile saline and then inoculated in brain heart infusion broth. Likewise, pus and ear swab were enriched in brain heart infusion broth. Broth cultures were incubated at 37^0C for 24 hrs and examined for morphology of organism by Gram's staining. Based on morphological characteristics, broth cultures were further processed for isolation of specific bacteria.

Isolation and identification of bacteria
Standard protocol was followed for isolation and identification of bacteria (Bergey *et al.*, 1994). Broth culture showed presence of both Gram positive cocci and Gram negative bacilli. Broth culture were streaked on to nutrient agar, mannitol salt agar, 10% sheep blood agar, eosine methylene blue (EMB) agar and Mac Conkey Lactose agar (MLA) and incubated at 37^0C for 24 hrs. Isolates were then subjected to series of biochemical tests *viz.* tube coagulase, catalase, oxidase, motility, indole production, methyl red (MR), Voges Proskauer (VP), citrate utilization, 1% maltose utilization in purple agar, urease and H_2S production in triple sugar iron agar (TSI) slant.

Antibiotic sensitivity test
The antibiotic sensitivity test was conducted by the disc diffusion method as per Bauer *et al.* (1966) [3] in Muller Hinton agar. Antibiotic discs (Himedia) of commercially available antimicrobials such as penicillin-G (10 IU), ampicillin (10µg), amoxycillin (30µg), amoxyclav (30µg), trimethoprim (10µg), chloramphenicol (30µg), ciprofloxacin (5µg), enrofloxacin (10µg), cefoxitin (30µg), cefepime (30µg), ceftazidime (30µg), ceftriaxone (30 µg), ceftazidime/ tazobactum (30/10 µg), ceftriaxone/tazobactum (30/10 µg), imipenem-EDTA (10/750 µg), gentamicin (10µg), amikacin (30 µg), tetracycline (30µg), doxycycline (30µg) and azithromycin (30 µg). The diameter of zone of inhibition was measured and categorized as susceptible, intermediate or resistant.

Results and Discussion
Bacterial isolates
Bacterial isolates *viz. Pseudomonas aeruginosa*, *Staphylococcus aureus* and *E. coli* were isolated and identified from clinical specimens on the basis of cultural and biochemical characteristics.
Pseudomonas aeruginosa was identified on the basis of cultural characteristics including smooth, mucoid colonies with grape like odour and greenish pigmentation on nutrient agar, haemolysis on blood agar and non lactose fermentation on MLA. *Pseudomonas aeruginosa* isolates were motile and showed positive reaction for catalase, oxidase, urease and citrate utilization where as negative for MR, indole and VP. Bacterial isolates showing golden yellow pigmented colony on nutrient agar, double haemolysis on blood agar and pink colony on MLA were identified as *Staphylococcus aureus*. *Staphylococcus aureus* isolates were positive for catalase, VP, MR, urease, maltose utilization and negative for indole, oxidase, H_2S and were non-motile. Isolates producing pink, mucoid lactose fermenting colony on MLA and metallic sheen over EMB agar were identified as *E. coli* and further confirmed by IMViC test. *E.coli* isolates were nonhemolytic except five isolates obtained from UTI. All *E. coli* isolates were motile, catalase, indole and MR positive and were negative for oxidase, VP, H_2S, urease and citrate utilization.

Prevalence of bacterial isolates
Predominant isolates in UTIs was E. coli followed by *Staphylococcus aureus* and *Pseudomonas aeruginosa* during both the course of study whereas in pyoderma major isolates were *Staphylococcus aureus* followed by *Pseudomonas aeruginosa* and *E. coli* (Table 1 and 2).

Table 1: Bacterial isolates from dogs during 2013-14

Name of bacterial isolates	Number of bacterial isolates			
	UTI	Chronic recurrent pyoderma	Otitis externa	Total
Staphylococcus aureus	10(34.48%)	12 (48%)	9 (40.91%)	31 (40.79%)
E. coli	13 (44.83%)	6 (24%)	6 (27.27%)	25 (32.89%)
Pseudomonas aeruginosa	6 (20.69%)	7 (28%)	7 (31.82%)	20 (26.32%)
Total	29	25	22	76

Table 2: Bacterial isolates from dogs during 2016-17

Name of bacterial isolates	Number of bacterial isolates			
	UTI	Chronic recurrent pyoderma	Otitis externa	Total
Staphylococcus aureus	12 (32.43%)	13 (40.63%)	6 (28.57%)	32 (35.16%)
E. coli	17 (45.95%)	7 (21.88%)	5 (23.81%)	29 (31.87%)
Pseudomonas aeruginosa	8 (21.62%)	12 (37.50%)	10 (47.62%)	30 (32.97%)
Total	37	32	21	91

In cases of otitis externa variation in prevalence was noted during two different course of study; *Staphylococcus aureus* being predominant during 2013-14 and *Pseudomonas aeruginosa* during 2016-17 as showing in Table 1 and 2. It is also observed from the study that occurrence of *Pseudomonas aeruginosa* in clinical samples got increased in 2016-17 when compared with 2013-14 reports.
In accordance with the present findings, Roopali *et al.* (2018) [24] and Seguin *et al.* (2003) [29] reported *E. coli* as predominant pathogen in UTI of dogs. Similarly, Norris *et al.* (2000) [18] and Ball *et al.* (2008) [2] reported *Staphylococcus* spp. as the second or third most common isolate in UTI of dogs. Existence of *Pseudomonas aeruginosa* in recurrent pyoderma in dog is also supported by previous finding of Sannat *et al.* (2014) [26].

Trends in antibiotic resistance
Antibiotic sensitivity pattern of bacterial isolates *i.e. Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E. coli* are shown in Table 3 to 5.

Table 3: Trends in antibiotic resistance of *Staphylococcus aureus*

Name of antibiotic	Number(%) of sensitive *Staphylococcus aureus* isolates	
	2013-14 (31isolates)	2016-17 (32 isolates)
Imipenem	31(100%)	31(96.88%)
Cefepime	30(96.77%)	27(84.38%)
Cefoxitin	30(96.77%)	28(87.5%)
Ceftazidime/tazobactum	29(93.55%)	27(84.38%)
Ceftriaxone tazobactum	29(93.55%)	26(81.25%)
Azithromycin	28(90.32%)	24(75%)
Doxycycline	26(83.87%)	22(68.75%)
Ciprofloxacin	25(80.65%)	26(81.25%)
Chloramphenicol	23(74.19%)	25(78.12%)
Amoxyclav	23(74.19%)	16(50%)
Gentamicin	23(74.19%)	17(53.13%)
Enrofloxacin	21(67.74%)	20(62.5%)
Ceftriaxone	21(67.74%)	14(43.75%)
Amikacin	21(67.74%)	21(65.63%)
Ceftazidime	20(64.51%)	12(37.5%)
Tetracycline	19(61.29%)	18(56.25%)
Amoxycillin	15(48.39%)	10(31.25%)
Ampicillin	13(41.94%)	12(37.5%)
Trimethoprim	3(9.68%)	0 (0%)
Penicillin-G	2(6.45%)	0(0%)

Table 4: Trends in antibiotic resistance of *E. coli*

Name of antibiotic	Number(%) of sensitive *E. coli* isolates	
	2013-14 (25 isolates)	2016-17 (29 isolates)
Imipenem	25(100%)	28(96.55%)
Azithromycin	25(100%)	26(89.66%)
Cefoxitin	25(100%)	24(82.76%)
Cefepime	23(92%)	26(89.66%)
Ceftazidime/tazobactum	24(96%)	25(86.20%)
Chloramphenicol	24(96%)	26(89.66%)
Ciprofloxacin	24(96%)	19(65.52%)
Ceftazidime	24(96%)	27(93.10%)
Amikacin	24(96%)	26(89.66%)
Ceftriaxone tazobactum	23(92%)	22(75.86%)
Doxycycline	23(92%)	24(82.76%)
Gentamicin	23(92%)	21(72.41%)
Enrofloxacin	22(88%)	21(72.41%)
Ceftriaxone	21(84%)	19(65.52%)
Tetracycline	19(76%)	19(96.55%)
Trimethoprim	19(76%)	18(62.07%)
Amoxyclav	15 (60%)	12(41.37%)
Amoxycillin	14(56%)	12(41.37%)
Ampicillin	13(52%)	12(41.37%)

Table 5: Trends in antibiotic resistance of *Pseudomonas aeruginosa*

Name of antibiotic	Number(%) of sensitive *Pseudomonas aeruginosa* isolates	
	2013-14 (20 isolates)	2016-17 (30 isolates)
Imipenem	18(90%)	23(83.33%)
Azithromycin	16(80%)	19(63.33%)
Cefepime	16(80%)	21(70%)
Ceftazidime/tazobactum	15(75%)	18(60%)
Cefoxitin	15(75%)	19(63.33%)
Doxycycline	15(75%)	18(60%)
Amikacin	15(75%)	18(60%)
Ceftriaxone tazobactum	14(70%)	16(53.33%)
Gentamicin	14(70%)	19(63.33%)
Ciprofloxacin	13(65%)	12(40%)
Enrofloxacin	10(50%)	10(33.33%)
Chloramphenicol	13(65%)	16(53.33%)
Ceftriaxone	13(65%)	12(40%)
Tetracycline	12(60%)	16(53.33%)
Ceftazidime	10(50%)	13(43.33%)
Amoxyclav	9(45%)	5(16.66%)
Ampicillin	9(45%)	8(26.66%)
Amoxycillin	7(35%)	6(20%)
Trimethoprim	5(25%)	7(23.33%)

During course of present investigation multidrug resistant *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *E. coli* isolates were reported from UTI, otitis externa and recurrent pyoderma of dog. Apparently, increasing trends in antibiotic resistance were recorded against all three bacterial isolates. Though all bacterial isolates under study were least resistant to imipenam but emergence of imipenam resistant isolates were also reported during the year 2016-17. There was substantial increase in resistance of *Staphylococcus aureus* isolates for amoxyclav, amoxicillin, ceftazidime, ceftriaxone and gentamicin during 2016-17 when compared with the year 2013-14 whereas isolates showed increased sensitivity to ciprofloxacin and chloramphenicol. Considerable increase in antibiotic resistance among *E. coli* isolates were noted for ciprofloxacin, amoxyclav, amoxicillin and ceftriaxone during 2016-17 as compared with 2013-14. Similarly there was a drastic increase in antibiotic resistance among *Pseudomonas aeruginosa* for antibiotics namely ciprofloxacin, amoxyclav, imipenam, azithromycin, ampicillin and ceftriaxone.

Resistance pattern of *Staphylococcus aureus*

Most of the *Staphylococcus aureus* isolates were sensitive to imipenem, cefepime, cefoxitin, ceftazidime tazobactum, ceftriaxone /tazobactum and azithromycine whereas timethoprim and penicillin-G were least effective. Staphylococci, besides being commensals on mucosal surfaces and skin, are often implicated in a variety of infections, as otitis externa, abscesses, furuncles, pyoderma, pneumonia, and bacteremia in dogs, in both veterinary hospitals and the general population, and can be transmitted readily between animals and human (Lilenbaum *et al.*, 2003) [12]. Members of this genus have a high frequency of conjugation and frequently acquire plasmids that encode antimicrobial resistance (Malik *et al.*, 2005) [16].

Likewise present observation, Vincze *et al.* (2010) also described multidrug resistant *Staphylococcus* as a cause of canine pyoderma. Earlier reports of MDR Staphylococci in UTI of (Joseph and Matthew, 2011 and Wong *et al.* 2015) and horse (Shende *et al.* 2016) supports the present findings. In their study, they reported that isolates were resistant to various antibiotics including ampicillin, methicillin, penicillin, ceftiofur, clindamycin, trimethoprim, gentamicin, enrofloxacin and cefoxitin. Present report is also in corroboration with findings of Tomar *et al.* (2015) who observed *Staphylococcus aureus* resistant to cefepime, gentamicin, ciprofloxacin, penicillin, ampicillin and amoxicillin. The resistance to methicillin (MRSA) confers resistance to all β-lactam drugs, rendering many of the common treatment options for staphylococcal infections ineffective (Palavecino, 2007).Earlier reports confirming tetracycline (Benito *et al.*, 2016) and cefoxitin resistance as a marker of the livestock-associated MRSA supports existence of MRSA in present case (Fernandes *et al.*, 2005). In concordance with present findings, Couto *et al.* (2016) studied 16 years trends in antibiotic resistance of *Staphylococcus aureus* from clinical samples of horses and dogs and reported significant increase in antimicrobial resistance and occurrence of MRSA isolates. Several clonal lineages of MRSA circulating in human hospitals and the community were found, suggesting that companion animals can become infected with and contribute to the dissemination of highly successful human clones (Couto *et al.*, 2016).

Resistance pattern of *Pseudomonas aeruginosa*

Though most of the isolates of *Pseudomonas aeruginosa* were sensitive to imipenam but each isolates showed resistant to at least 2 or 3 antibiotics. *Pseudomonas aeruginosa* was reported as most drug resistant pathogens as compared with *E. coli* and *Staphylococcus aureus. Pseudomonas aeruginosa* is clinically important bacteria as it can survive in conditions that few other organisms can tolerate and they also produce slime layer to defend against phagocytosis in the bloodstream of the host (Stolp and Gadkari, 1981).The relative sensitivity of *Pseudomonas aeruginosa* to imipenem observed in the present study is in concordance with findings of Mansour *et al.* (2013) who also reported sensitivity to imipenam and resistance to ceftazidime and ciprofloxacin. Similarly, Haleem *et al.* (2011) reported *Pseudomonas* isolates resistant to cefotaxim, chloramphenicol, penicillin and ampicillin and moderately sensitive to azithromycin and gentamicin. Likewise Tomar *et al.* (2015) and Sannat *et al.* (2014) reported *Pseudomonas aeruginosa* resistant to cefepime, gentamicin, enrofloxacin, doxycycline, chloramphenicol and ciprofloxacin. Existance of MDR *Pseudomonas aeruginosa* is also supported by Loureiro *et al.* (2002). This resistance might be due to presence of β--lactamase enzymes or due to decreased membrane permeability towards antimicrobial agents (Al-Falahy, 2000). Likewise present study, Sharma et (2016) in a retrospective analysis observed that the lowest percentage of resistance manifested against imipenem was 14%, 24.48% and 20.99% for the years 2013-2015 and resistance for fluoroquinolones decreased over the three consecutive years from 84.67% (2013) to 78.27% (2015); multi drug resistant pseudomonas percentage resistant to fluoroquinolones, third generation cephalosporines and aminoglycosides were 78% (2013), 78.33% and 80.33% (2015) over the consecutive years.

Resistance pattern of *E. coli*

In first phase of study *E. coli* isolates were 100% sensitive to imipenem, azithromycin and cefoxitin however emergence of resistance was recorded in second phase. Most of the antibiotic was found effective against *E. coli* except amoxyclav however increasing trend of resistance were noted to each antibiotic tested. *E. coli* was the most common pathogen isolated from the canine UTI which is consistent with observations of Ling *et al.* (2001), Hall *et al.* (2013) and Wong *et al.* (2015). In concordance with the findings of present study, Wong *et al.* (2015) detected MDR *E. coli* in UTI of dogs and observed resistance to amoxyclav, ampicillin, chloramphenicol, doxycycline, enrofloxacin and trimethoprim. Similarly, Sarah and Ann (2013) reported non-haemolytic *E. coli* from ulcerative cystitis which was resistant to sulfamethoxazole/trimethroprim, ampicillin–sulbactam, and tetracycline. Kevin *et al.* (2015) demonstrated higher frequency of resistance to cephalosporins than to any other class of agent and nearly 20% *E.coli* isolates resistant to enrofloxacin. Resistance to amoxyclav and fluoroquinolones in *E.coli* has also been reported by Weese *et al.* (2011) and Wanger *et al.* (2014). Various reports have documented the emergence of *E. coli* capable of producing broad-spectrum β-lactamases and it is of immense significance since the β-lactam antimicrobials are of therapeutic importance in humans and many domestic animals. Selection pressure associated with prior antimicrobials therapy might be responsible for the relatively high resistance to cephalosporins and β-lactamase genes in uropathogenic *E. coli* (Pitout, 2012). Increased resistance trends of *E. coli* against aminoglycosides, fluoroquinolone and ceftriaxone over a period of three years was reported by Sharma *et al.* (2016) in Punjab, which more

or less corresponds to present observation.
Though the sample size was relatively less in present investigation, it present important data pertaining to regional trends and patterns of antimicrobial resistance of MDR pathogens associated with clinical cases not responding to conventional antibiotic therapy.

Conclusion
Present study reports existence of multidrug resistance *Staphylococcus aureus, E. coli* and *Pseudomonas aeruginosa* from UTI, otitis externa and recurrent pyoderma of dog. Increasing trends in antibiotic resistance were recorded against these bacterial isolates. Bacterial isolates were found resistant to most of the antibiotics tested except imipenam. Emergence of resistance to new generation antibiotics like imipenam and cefoxitin is a alarming situation for both human and animal health because transmission of drug resistance occurs by direct contact between animals and human. Hence, appropriate legislation is needed to support the responsible and prudent use of antimicrobial agents in veterinary medicine to prevent transmission of drug resistance to human beings.

References

1. Al-Falahy RN. Bacteriological study on some isolates of *Pseudomonas aeruginosa* resistant to antibiotics and study plasmid content. M.Sc. Thesis submitted to Al-Mustansiryia University, 2000.
2. Ball KR, Rubin JE, Chirino-Trejo M. Antimicrobial resistance and prevalence of canine uropathogens at the Western College of Veterinary Medicine Veterinary Teaching Hospital, 2002–2007. Can. Vet. J. 2008; 49:985-990.
3. Bauer AW, Kirby WMM, Shernis JC, Turck M. Antibiotic susceptibility testing by standardized single disc method. Am. J Clin. Pathol. 1966; 45:493-497.
4. Benito D, Gomez P, Aspiroz C, Zarazaga M, Lozano C, Torres T. Molecular characterization of *Staphylococcus aureus* isolated from humans related to a livestock farm in Spain, with detection of MRSA-CC130 carrying mec C gene: A zoonotic case? Enferm. Infecc. Microbiol. Clin. 2016; 34(5):280-285.
5. Bergey DH, Holt JG, Krieg NR, Sneath PHA. Bergey's Manual of Determinative Bacteriology, 9th edn. (Breed RS, Murray EGD and Smith NR, Eds.) Williams and Wilkims, Baltimore, 1994.
6. Couto N, Monchique C, Belas A, Marques C, Gama LT, Pomba C. Trends and molecular mechanisms of antimicrobial resistance in clinical staphylococci isolated from companion animals over a 16 year period. J Antimicrob. Chemother. 2016; 71(6):1479-87.
7. Fernandes CJ, Fernandes LA, Collignon P. Cefoxitin resistance as a surrogate marker for the detection of methicillin-resistant Staphylococcus aureus. J. Antimicrob. Chemother. 2005; 55:506-510.
8. Haleem H, Tarrad JK, Banyan IA. Isolation of *Pseudomonas aeruginosa* from Clinical Cases and Environmental Samples, and Analysis of its Antibiotic Resistant Spectrum at Hilla Teaching Hospital. Med. J Babylon. 2011; 8(4):34-41.
9. Hall JL, Holmes MA, Baines SJ. Prevalence and antimicrobial resistance of canine urinary tract pathogens. Vet. Rec. 2013; 173:549.
10. Joseph ER, Matthew CG. Urinary tract infection caused by methicillin-resistant *Staphylococcus pseudintermedius* in a dog. Can. Vet. J. 2011; 52:162-164.
11. Kevin JC, Victor AA, Craig A. Antimicrobial resistance trends among canine *Escherichia coli* isolates obtained from clinical samples in the northeastern USA, 2004-2011. Can. Vet. J. 2015; 56(4):393-398.
12. Lilenbaum W, Veras M, Blum E, Souza GN. Antimicrobial susceptibility of staphylococci isolated from otitis externa in dogs. Lett. Appl. Microbiol. 2003; 31:42-45.
13. Ling GV, Norris CR, Franti CE. Interrelations of organism prevalence, specimen collection method, and host age, sex, and breed among 8,354 canine urinary tract infections (1969–1995). J Vet. Intern. Med. 2001; 15:341-347.
14. Lloyd DH. Reservoirs of antimicrobial resistance in pet animals. Clin. Infect. Dis. 2007; 45(2S):148-152.
15. Loureiro MM, De-Moraes B, Mendonca VLF, Pinheiro GS, Asensi MD. *Pseudomonas aeruginosa*: Study of antibiotic resistance and molecular typing in hospital infection cases in a neonatal intensive care unit from Riode Janeiro City, Barzel. Members Institutes of Oswaldo Criz, Riode Janeiro. 2002; 97(3):387-394.
16. Malik S, Peng H, Barton MD. Antibiotic resistance in staphylococci associated with cats and dogs. J Appl. Microbiol. 2005; 99:1283-1293.
17. Mansour SA, Eldaly O, Jiman-Fatani A, Mohamed ML, Ibrahim EM. Epidemiological characterization of P. aeruginosa isolates of intensive care units in Egypt and Saudi Arabia. East. Mediterr. Health J. 2013; 19:71-80.
18. Norris CR, Williams BJ, Ling GV. Recurrent and persistent urinary tract infections in dogs: 383 cases (1969–1995). J. Am. Anim. Hosp. Assoc. 2000; 36:484-492.
19. Oluoch AO, Kim CH, Weisiger RM. Nonenteric *E. coli* isolates from dogs: 647 cases (1990-1998). Journal of American Veterinary Medicine Association. 2001; 172:708-711.
20. Palavecino E. Clinical, epidemiological, and laboratory aspects of methicillin-resistant *Staphylococcus aureus* (MRSA) infections. Methods Mol. Biol. 2007; 391:1-19.
21. Passmore CA, Sherington J, Stegemann MR. Efficacy and safety of cefovecin (Convenia) for the treatment of urinary tract infections in dogs. Journal of Small Animal Practice. 2007; 48(3):139-144.
22. Pitout JD. Extraintestinal pathogenic *Escherichia coli*: A combination of virulence with antibiotic resistance. Front Microbiol. 2012; 3:9.
23. Ramkumar Singh Tomar, Amit Kumar Gupta, Chandrahas Sannat, Ritu Agrawal, Hirpurkar SD. Assessment of Antimicrobial Activity of Pomegranate Leaf Extract against Bacterial Isolates of Otitis Externa and Diarrhea of Dogs. Journal of Animal Research. 2015; 5(4):767-772.
24. Roopali B, Roy M, Roy S. Haemato-biochemical changes and therapeutic management of urinary tract infection in canines. The Pharma Innovation Journal. 2018; 7(6):89-92.
25. Roy M, Kashyap R, Roy S. Prevalence of Canine Otitis Externa, Etiology and Clinical Practice in and around Durg District of Chhattisgarh State, India. Int. J Curr. Microbiol. App. Sci. 2018; 7(3):269-274.
26. Sannat C, Shende R, Dilliwar V, Hirpurkar SD. Studies on isolation and antibiogram profile of pseudomonas

aeruginosa from recurrent pyoderma of canine Veterinary practitioner. 2014; 15(1):38-39.
27. Sarah CS, Ann PB. An unusual case of urinary retention and ulcerative cystitis in a horse, sequelae of pelvic abscessation, and adhesions. Can. Vet. J. 2013; 54:690-692.
28. Seckerdieck F, Mueller RS. Recurrent pyoderma and its underlying primary diseases: a retrospective evaluation of 157 dogs Veterinary Record, 2018, 182-434.
29. Seguin MA, Vaden SL, Altier C, Stone E, Levine JF. Persistent urinary tract infections and reinfections in dogs (1989-1999). Journal of American Veterinary Medicine Association. 2003; 17:622-631.
30. Sharma N, Gupta A, Walia G, Bakhshi R. Pattern of Antimicrobial Resistance of Escherichia coli Isolates from Urinary Tract Infection Patients: A Three Year Retrospective Study. J Appl. Pharma. Sci. 2016; 6(1):062-065.
31. Stolp H, Gadkari D. *In:* The prokaryotes (Starr, P.M.; Stolp, H., Turner, H.G., Balawas, A., and Schegel, H.G. Eds.). 1981; 1:721-741.
32. Wagner S, Gally DL, Argyle SA. Multidrug-resistant *Escherichia coli* from canine urinary tract infections tend to have commensal phylotypes, lower prevalence of virulence determinants and amp C-replicons. Vet. Microbiol. 2014; 169:171-178.
33. Weese JS, Blondeau JM, Boothe D, Breitschwerdt EB, Guardabassi L, Hillier A *et al*. Antimicrobial use guidelines for treatment of urinary tract disease in dogs and cats: antimicrobial guidelines working group of the international society for companion animal infectious diseases. Vet. Med. Int., 2011. Article ID 263768 (1-9). doi: 10.4061/2011/263768.
34. Wong MH, Chan EW, Chen S. Evolution and dissemination of oqxAB- like efflux pumps, an emerging quinolone resistance determinant among members of Enterobacteriaceae. *Antimicrob.* Agents Chemother. 2015; 59:3290-3297.