Bacterial culture and antibiotics susceptibility in chronic suppurative otitis media at the secondary care hospital in North India

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Research Article

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Abstract

Background: Chronic suppurative otitis media (CSOM) is defined as a persistent infection of the middle ear with a perforated tympanic membrane and draining exudate for more than 6 weeks. Information about the organism responsible for COSM and their antibiotic susceptibility pattern is an important for effective treatment.

Aim: This study aims to develop protocol for empirical treatment by determining aerobic bacterial profile and antibiotics susceptibility in patients of chronic suppurative otitis media (CSOM) at a secondary care hospital in North India.

Material and Methods: A cross-sectional study was conducted at ENT department of the secondary care hospital in North India on 85 patients, middle ear discharge sample was collected under strict aseptic conditions and antibiotic susceptibility done as per Clinical Laboratory Standards Institute guidelines.

Result: 85 ear swabs were collected, and 89 bacterial isolates were identified, of which 62 (72.94%) sample with mono-microbial growth, 14 (16.47%) with polymicrobial growth, 8 (9.41%) show no growth and rest 1(1.17%) was contaminant. Among 89 isolates, 35 (39.33%) were Gram-positive bacteria, while 54 (60.67%) were Gram-negative bacteria. The most common isolates were *Pseudomonas spp*. (36; 40.45%), followed by *MSSA* (34; 38.20%), *Proteus spp*. (7; 7.87%), *Klebsiella spp*.(3; 3.37%), *Enterobacter spp*.(3; 3.37%), *E. coli* (3; 3.37%), *Actinobacteria spp*. (2; 2.25%) and *MRSA* (1; 1.12%). *Pseudomonas spp.* showed 100% susceptible to colistin, linezolid, imipenem, amikacin (97%); ciprofloxacin (92%); gentamicin (95%); Ceftriaxone (83%); meropenem (93%); Netilmicin (98%) and SXT (90%). Proteus spp. was 100% susceptible to amikacin, ciprofloxacin, Imipenem, meropenem, netilmicin; ampicillin (71%); amoxicillin–clavulanic acid (85%); ceftriaxone (85%); gentamicin (85%) and SXT (85%). Among Gram-positive bacteria, MSSA was 100 % susceptible to meropenem and Imipenem, amikacin (97%); gentamicin (81%); amoxicillin–clavulanic acid (91%); linezolid (92%); Netilmicin (94%); Vancomycin (91%); Colistin (97%) and SXT (41%). MRSA showed 100% susceptibility to gentamicin, netilmicin and vancomycin.

Conclusion: Pseudomonas and MSSA were the principal bacterial isolate responsible for causing CSOM in this study though the most common organism was Pseudomonas spp. We conclude the combination of amikacin and ceftriaxone to be used as systemic therapy

1. Introduction:

Chronic suppurative otitis media (CSOM) is defined as a persistent infection of the middle ear with a perforated tympanic membrane and draining exudate for more than 6 weeks.(Brennan-Jones et al., 2020) Thirty-one million people developed CSOM across the globe every year and with prevalence of disease is as much as three times higher in developing countries than western developed world, around the world. (World Health Organization, 2018) The World Health Organization (WHO) has categorized CSOM as neglected tropical diseases and is more common in low socioeconomical status.

CSOM usually occurs due to in poor and inadequate management of acute otitis media. The etiology and antimicrobial resistance patterns of CSOM infection are different in different geographical area and population studied.(Gebreyes et al., 2006)(Tripodi et al., 2005) CSOM is predominantly caused by Pseudomonas aeruginosa, Escherichia coli, S. aureus, Streptococcus pyogenes, Proteus mirabilis, Klebsiella species among aerobic bacteria. However, Bacteroides, Pepto streptococcus, Propionibacterium are common anaerobic bacteria.(Leung et al., 1998)

In India studies had been conducted at various parts of country to establish the common pathogen and their antibiotic susceptibility, some study concluded as Pseudomonas spp. other shows Staphylococcus spp. as most common aerobic bacterial isolate involved in CSOM. Some shows polymicrobial pathogens. (Keam et al., 2005) Anaerobic pathogens like *Peptostreptococcus, Bacteroides* and *Peptococcus* involved in 10-11% cases of CSOM.(Kucisec-Tepes et al., 2006) Most common fungal agent to be *Aspergillus spp* which was nearly 83% of all fungal causes of CSOM and second most common cause was *Candida spp.*

Thus, information about the organism responsible for COSM and their antibiotic susceptibility pattern is an important for effective treatment. Therefore, the aim of this study was to determine bacterial profile and their antibiotic susceptibility patterns in COSM from patients attending the ENT clinic of the hospital. This study aims to develop protocol for empirical treatment by determining aerobic bacterial profile and antibiotics susceptibility in patients of chronic suppurative otitis media (CSOM) at a secondary care hospital in North India. (Mukherjee et al., 2011)(Togano et al., 2018)

2. Material And Methods:

2.1. Study design and Setting:

A cross-sectional study was conducted at ENT department of the secondary care hospital in North India. The total of 85 patients with unilateral or bilateral active chronic otitis media studied. The study participants were enrolled consecutively using a convenience sampling technique. All study participants had perforated tympanic membranes with active purulent discharge. The detailed information regarding age, sex, duration of discharge, and the antibiotic is taken prior to pus collection was collected from each study participant using a structured questionnaire in the ENT OPD. Whereas ear discharge of less than 6 weeks duration, otitis externa, and patient taking topical/systemic antibiotic within 7 days before pus collection were excluded.

2.2. Sample collection:

Middle ear discharge sample was collected at ENT OPD under strict aseptic conditions using single use sterile culture swabs, after proper cleaning the external auditory canal using povidone iodine and spirit swab for each ear. The swabs were transported to the microbiology laboratory in the department of pathology of the same hospital for culture and antibiotic susceptibility testing.

2.3. Culture and identification:

Swab was directly used for inoculating on 5% sheep blood agar, MacConkey agar. The blood and MacConkey agar plates were incubated aerobically was incubated at 37 °C for 24–48 h. The isolates were

identified by colony morphology, Gram stain and standard microbiological tests like oxidase test, coagulase test and biochemical tests like triple sugar iron test, indole test, citrate utilization test and urease test. (Palù, 1999)

Antibiotic susceptibility tests were performed using a modified Kirby–Bauer disc diffusion method following the Clinical Laboratory Standards Institute guidelines.

2.4. Data analysis and interpretation:

Data were entered and analyzed using SPSS version 20 software. Results were presented through graphs and tables. The statistical significance of association was measured by using the Chi-square test. A p-value < 0.05 was considered as statistically significant.

3. Results:

A total of 85 patients participated in the study. Out of which 57 (67.05%) were male and 28 (32.94%) were female. The mean age of the study participant was 23 years and ranging from 4 to 68 years. Thirty-nine (45.88%) of them were aged below 20 years, 37 (43.57%) were between 21- 40 years, 7 (8.24%) were between 41- 60 years and 2 (2.35%) were above 61 years. 29 (34.11%) patients had bilateral disease, 56 (65.88%) of the patients had unilateral CSOM. Left ear disease seen in 33 (58.93%) patients.

Of the 85 ear swabs, 89 bacterial isolates were identified. Of which 62 (72.94%) sample with mono-microbial growth, 14 (16.47%) with polymicrobial growth, 8 (9.41%) show no growth and remaining 1 (1.17%) shows contaminants. Among 89 isolates, 35 (39.33%) were Gram-positive bacteria, while 54 (60.67%) were Gram-negative bacteria. The most common isolates were Pseudomonas spp. (36; 40.45%), followed by MSSA (34; 38.20%), Proteus spp. (7; 7.87%), Klebsiella spp.(3; 3.37%), Enterobacter spp.(3; 3.37%), E. coli (3; 3.37%), Actinobacteria spp. (2; 2.25%) and MRSA (1; 1.12%).

Mixed cultures (14;16.47%) seen in equal percentage in both adult and child population, with a combination of Pseudomonas and MSSA in 50% (7/14); Enterobacter spp. and MSSA 14.29% (2/14); Acinetobacter spp./ E. coli/ Proteus spp. and MSSA 7.14% (each 1/14); Proteus spp. and Pseudomonas spp. 7.14% (1/14) and Acinetobacter spp. and Enterobacter spp. 7.14% (1/14). Antibiotic susceptibility patterns of most isolated Gram-negative and Gram-positive bacteria, respectively. Pseudomonas spp. showed 100% susceptible to colistin, linezolid, imipenem; amikacin (97%); ciprofloxacin (92%); gentamicin (95%); Ceftriaxone (83%); meropenem (93%); Netilmicin (98%) and SXT (90%). Proteus spp. was 100% susceptible to amikacin, ciprofloxacin, Imipenem, meropenem, netilmicin; ampicillin (71%); amoxicillin–clavulanic acid (85%); ceftriaxone (85%); gentamicin (85%) and SXT 985%). Among Gram-positive bacteria, MSSA was 100 % susceptible to meropenem and Imipenem; amikacin (97%); gentamicin (81%); amoxicillin–clavulanic acid (91%); linezolid (92%); Netilmicin (94%); Vancomycin (91%); Colistin (97%) and SXT (41%). MRSA showed 100% susceptibility to gentamicin, netilmicin and vancomycin.

The resistance pattern among the gram-positive isolates were, MRSA was 100 % resistance to ampicillin, linezolid and SXT. (Figure 1) MSSA was found resistance to ampicillin (65%) and SXT (59%). (Figure 2) In gram-negative isolates Pseudomonas spp. found resistance to Ampicillin (100%), amoxicillin–clavulanic acid (97%), ceftriaxone (47%) and SXT (28%);(Figure 3) Proteus spp. found resistance to Colistin (86%) and ampicillin (29%);(Figure 4) Klebsiella spp. found resistance to Colistin (100%), ampicillin (67%) and SXT (33%);(Figure 5) Enterobacter spp. and Acinetobacter spp. found 100% resistant to ampicillin;(Figure 6,7) E Coli. was 33% resistant to. amoxicillin–clavulanic acid, ciprofloxacin, and ceftriaxone. (Figure 8) (Table 1,2)

4. Discussion:

CSOM is defined as chronic otorrhea of more than 6 weeks duration through a perforated tympanic membrane. CSOM is a disease of multiple etiologies and is well known for persistence and recurrence despite treatment. The disease has irreversible sequalae and leads to serious intra and extra cranial complications due to proximity of various important structures viz facial nerve, auditory labyrinth, lateral sinus and middle and posterior fossa dura. CSOM is mainly classified as attico-antral (squamous COM) and tubo-tympanic (mucosal) type. According to world health organization (WHO) the prevalence of CSOM in Asian countries is about 4% which is much more than the developed western world.(World Health Organization, 2015) The indiscriminate use of antimicrobials in Asian countries lead to multiple drugs resistance among the infective organisms leading to treatment challenges. There are no gender predilections though the most surveys were done on children, CSOM can affect both the children as well the adults, as presented in this study.

Many researchers had addressed the issue for finding the common isolates and their sensitivity pattern among the antimicrobials in line of developing the treatment protocols in different regions of world. (Addis et al., 2011)(Saravanan and Raveendaran, 2013)

In present study 62 (72.94%) sample with mono-microbial growth, 14 (16.47%) with polymicrobial growth, 8 (9.41%) show no growth and remaining 1 (1.17%) shows contaminants. Our study is corelated with Gopichand WR et al and Tadesse et al in which most of the isolates are pure culture rather than mixed one.

The aerobic bacterial isolates in our study are *Pseudomonas spp.* (40.45%), followed by *MSSA* (38.20%), *Proteus spp.* (7.87%), *Klebsiella spp.*(3.37%), *Enterobacter spp.*(3.37%), E. coli (3.37%), *Actinobacteria spp.* (2.25%) and *MRSA* (1.12%). In our study the most common isolate was *Pseudomonas spp.* and it was like other studies, where also *Pseudomonas spp.* predominantly found in 48-98% of patients.

Imipenem was 100% sensitive to all gram-negative isolates consistent with the study of Rahim et al. (Yan et al., 2004) Amikacin was 100% sensitive in gram negative isolates except 8.33% of pseudomonas spp. and 2.94% samples of MSSA shows resistance to amikacin, this study is also at par of observation done by Maji et al. (Zhanel et al., 2008)(Pastagia et al., 2011) SXT was most effective antibiotic against MRSA as shown by Park et al in contrary to our study shows 100% resistance of MRSA spp. for SXT though it was not statistically significant.(Table1,2)

In general, our result has demonstrated that amikacin, ciprofloxacin, gentamicin, colistin, linezolid, imipenem, ceftriaxone are very sensitive to gram negative organism. Among Gram-positive bacteria meropenem, Imipenem, amikacin, gentamicin, amoxicillin–clavulanic acid, linezolid, Netilmicin, Vancomycin and Colistin are had bactericidal effects in majority.

The small sample size of this study may restrict the value of our finding, the frequently used antimicrobials in CSOM like penicillin developed high levels of resistance among the organisms like Pseudomonas and MSSA leading to treatment failures.

The CSOM still one of the most common cause of hearing impairment in India and the patients attending the ENT clinics have most frequent complaints of ear discharge with hard of hearing shows the morbidity among population due to CSOM. Though the main stay of treatment of CSOM is still ear drops (Quinolones, Aminoglycosides, chloramphenicol, neosporin and polymyxin-B) along with antihistamines and decongestants. The systemic therapy may be required in severe cases like complications or postoperative reasons.

5. Conclusion:

Pseudomonas and MSSA were the principal bacterial isolate responsible for causing CSOM in this study though the most common organism was *Pseudomonas spp*. We conclude the combination of amikacin and ceftriaxone to be used as systemic therapy and quinolones (ciprofloxacin) as topical therapy in CSOM.

Abbreviations:

MSSA: Methicillin sensitive staphylococcus aureus; MRSA: Methicillin resistant staphylococcus aureus; CSOM: chronic suppurative otitis media; ENT: ear, nose and throat; MDR: multidrug resistance. AMK- Amikacin, AMP- Ampicillin, AMC- amoxicillin–clavulanic acid, CIP- Ciprofloxacin, CRO- Ceftriaxone, CIN- Gentamicin, CT- Colistin, LZD- Linezolid, IMP- imipenem, MEM- Meropenem, NET- Netilmicin, SXT- Sulfamethoxazole and Trimethoprim and VAN- Vancomycin, ND- Not done

Tables

Table-1 Antimicrobial resistance in Gram-Negative Organism:

Isolates	Antibiotic Resistance (Gram Negative)												
	AMK n (%)	AMP n (%)	AMC n (%)	CIP n (%)	CRO n (%)	CIN n (%)	CT n (%)	LZD n (%)	IMP n (%)	MEM n (%)	NET n (%)	SXT n (%)	VAN n (%)
Pseudomonas spp.(n=36) Resistance (R)	3(8.33)	36(100)	35(96.7)	8(21.74)	17(47.22)	5(1.39)	0(0)	0(0)	0(0)	7(19.44)	2(5.55)	10(27.78)	ND
Proteus spp. (n=7) Resistance (R)	0	2(28.57)	1(14.9)	0	1(14.9)	1(14.9)	6(85.71)	ND	0	0	0	1(14.9)	ND
Klebsiella spp. (n=3) Resistance (R)	0	2(66.66)	0	0	0	ND	3(100)	ND	0	ND	ND	1(33.33)	ND
Enterobacter spp. (n=3) Resistance (R)	0	3(100)	3	0	0	0	0	ND	0	0	0	0	ND
Acinetobacter spp. (n=2) Resistance (R)	0	2(100)	0	0	1(50)	0	0	ND	0	0	ND	2(100)	ND
E. coli (n=3) Resistance (R)	0	0	1(33.33)	1(33.33)	1(33.33)	0	0	ND	0	0	ND	0	ND

Table-2 Antimicrobial resistance in Gram -positive Organism:

Isolates	Antibiotic Resistance (Gram Positive)												
	AMK n (%)	AMP n (%)	AMC n (%)	CIP n (%)	CRO n (%)	CIN n (%)	CT n (%)	LZD n (%)	IMP n (%)	MEM n (%)	NET n (%)	SXT n (%)	VAN n (%)
MRSA (n=1) Resistance (R)	ND	1 (100)	ND	0	ND	0	ND	1(100)	ND	ND	1(100)	1(100)	0
MSSA (n=34) Resistance (R)	1(2.94)	23(74.19)	3(8.82)	22(64.71)	ND	6(19.35)	1(2.94)	3(8.82)	0	0	2(5.88)	20(58.82)	3(8.82)

Declarations

Acknowledgement:

We would like to thank the study participants and department of microbiology for conducting the study.

Ethics Approval:

The ethical clearance for conducting the study was obtained from ESIC Model hospital, Noida. The patient and their guardian (in case of minor) was informed about the study and consent was taken.

Declaration of competing Interest:

Authors declare no conflict of interest.

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Figures

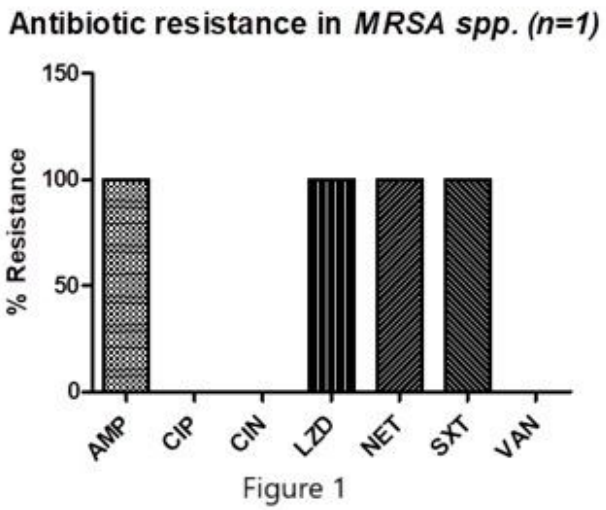

Figure 1

Figure 1

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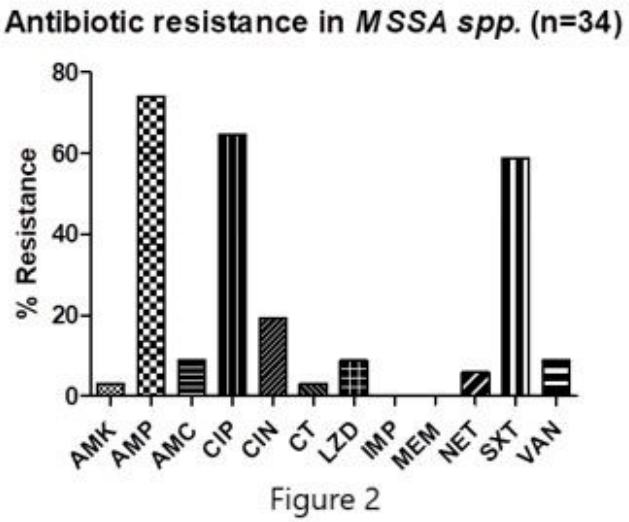

Figure 2

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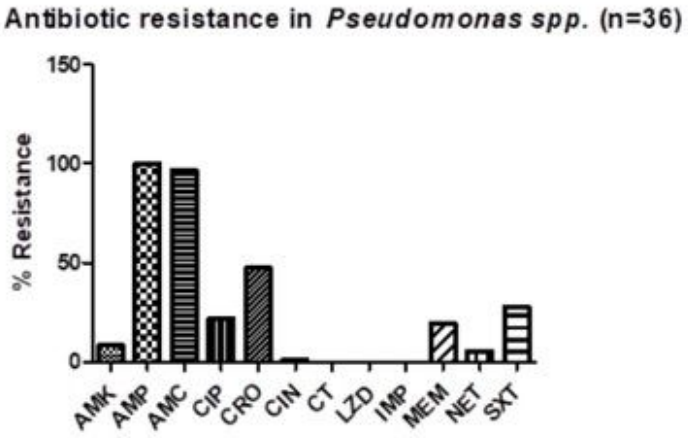

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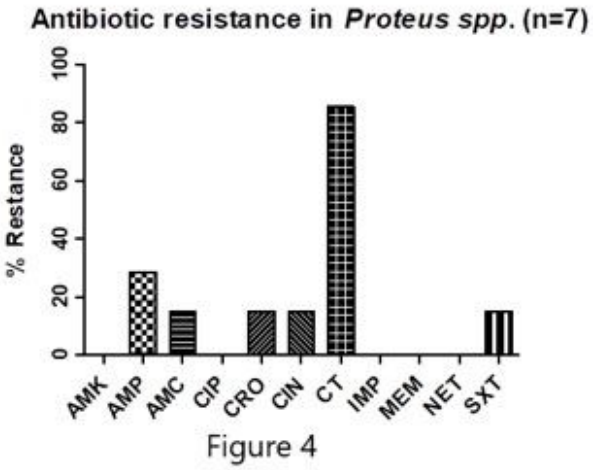

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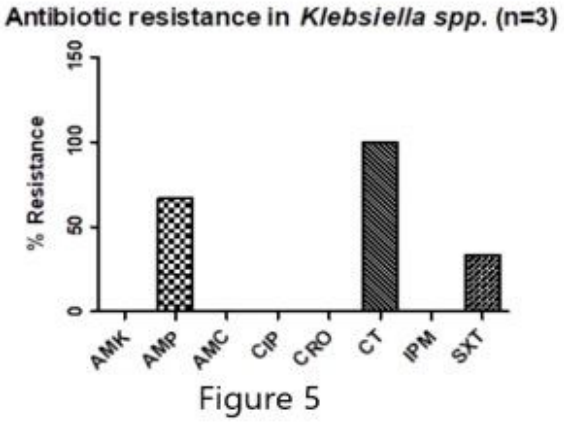

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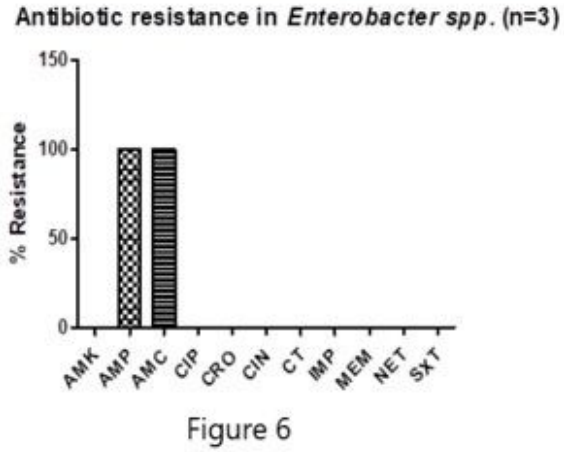

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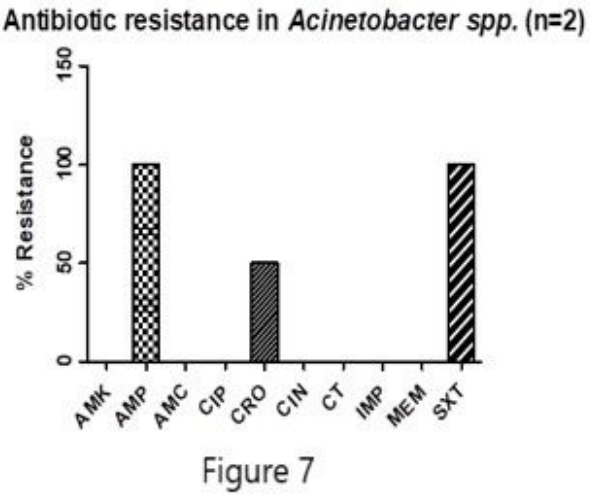

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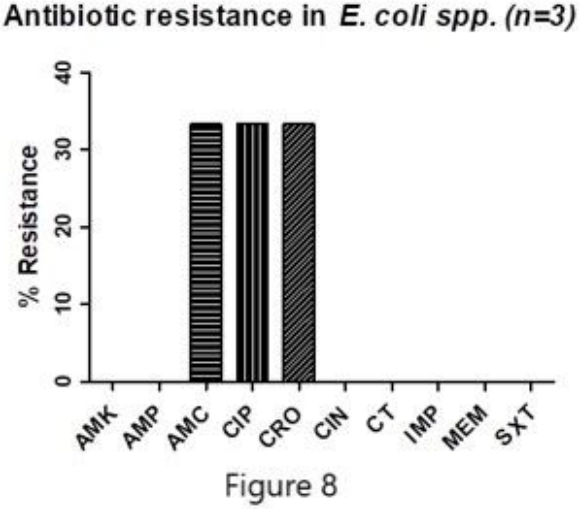

Figure 8

Figure 8