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Microbiological Profile and Antibiogram in Cases of Chronic Suppurative Otitis Media at a Tertiary Care Hospital, Jaipur, India

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A B S T R A C T

Otitis media is an inflammation of the middle ear cleft without reference to etiology or pathogenesis. Otitis media is more common in children, as their Eustachian Tube is shorter, narrower and more horizontal than the adult ear. Therefore, the study of microorganisms commonly associated with CSOM and their antibiotic sensitivity pattern is vital for the clinician to plan a general outline of treatment for the patient with a chronically discharging ear. Hence the aim of our study is to find out the fungal and bacterial aetiology and their antibiotic susceptibility pattern in clinically diagnosed CSOM cases in tertiary care hospital at Jaipur. The present study was carried on total of 53 clinically diagnosed CSOM cases attending ENT Department. After proper sample collection by sterile cotton swabs, they were immediately sent to the microbiology laboratory for aerobic bacterial culture, isolation and identification Conventional bacteriological and Mycological methods were used for identification of CSOM and susceptibility testing was performed as per CLSI guidelines 2017. This study comprises of 45 isolates of CSOM from total of 53 clinical specimens, collected from in- patients attended to ENT department during a period of 6 months. The most common organism causing CSOM among aerobic bacteria were *Pseudomonas species i.e.* 16 (29%) then *Staphylococcus aureus* 11(20%). In fungi most common organism isolated was *Aspergillus niger* then *Candida albicans*. Meropenem was most effective 100% against gram negative bacilli. In *Pseudomonas* Ticarcillin/Clavulanic Acid, Ceftazidime, Colistin and Polymyxin-B were 100% sensitive. Linezolid, Vancomycin and Rifampicin were 100% effective against gram positive cocci. The periodic evaluation of microbiological pattern and their antibiotic sensitivity pattern in local area becomes important and helpful in prescribing empirical antibiotics for successful treatment of CSOM and thus minimizing its complications and emergence of resistance strains.

Introduction

Otitis media is an inflammation of the middle ear cleft without reference to etiology or pathogenesis (Agrawal *et al.*, 2013). Otitis media is more common in children, as their Eustachian tube is shorter, narrower and more horizontal than the adult ear; and children, are at this age more prone to frequent upper respiratory tract infections which ascends to middle ear and may subsequently progress to CSOM. The difference in anatomy of the

paediatric ET favours creation of stasis of nasopharyngeal secretions making their movement difficult favouring the growth of microorganism than in adults (Bluestone *et al.,* 1988).

The acute form is acute suppurative otitis media usually associated with the infection in the upper respiratory tract whereas persistent form is known as chronic suppurative otitis media (CSOM) (Berman *et al.,* 1995) (Bluestone *et al.,* 1988).

Acute Suppurative otitis media is an acute inflammation of middle ear by pyogenic organism and is more common especially in infants and children of lower socioeconomic group (Dhingra *et al.,* 2004).

Chronic Suppurative Otitis Media is characterized by drainage from the middle ear for at least two weeks and is associated with a tympanic membrane perforation that is usually painless (Dhingra *et al.,* 2004).

The disease is mainly classified into two types (1) Mucosal or Tubotympanic type affecting the middle ear mucosa also called as 'safe disease', as it is characterized by a central perforation of the para tensa. (2) Squamosal or Atticoantral type which is an active squamous disease in which squamous epithelium present in middle ear cleft erodes the bone.

Bone erosion, with potentially dangerous results, was an inherent pathological features. Another synonym has been erosive middle ear disease and also called as 'Unsafe disease'.

The incidence of CSOM is higher in developing countries, especially among the low socioeconomic status of the society, use of poor nutrition, improper hygiene and lack of health education (Kumar *et al.,* 2011). The disease is highly prevalent in tropical regions including south Asia (Parveen *et al.,* 2012).

CSOM has direct impact on the hearing of patient causing conductive and sensorineural hearing loss and also on child development. It is also found to be the single major cause for conductive deafness, and is responsible for 1.5% speech disorders. Hearing loss associated with CSOM hampers educational skills in children that are well recognized by otologists, paediatricians and educators (Dhingra *et al.,* 2004).

The most common organism isolated now a days are *Pseudomonas aeruginosa, Staphylococcus aureus, Streptococcus pyogenes, Proteus species, Klebsiella species* among aerobes and among anaerobes *Bacteriodes, Peptostreptococcus, Proprionibacterium* (Barobby *et al.,* 1987).

Most common fungi *are Aspergillus species* and *Candida species* (Park *et al.,* 2008).

The diagnosis of COM is based on history, examination findings on otoscopy, supplemented by culture of the ear discharge and radiology of the temporal bone. Gram stains and cultures will assist in guiding therapy and are usually reserved for cases that fail standard topical therapy.

Computed tomography (CT) scan should be performed if extra cranial complications are suspected. CT scan has a role in diagnosis of COM when the TM cannot be visualized, for example by narrowing or stenosis of the EAC. It may reveal bone erosion from a cholesteatoma or point to area of potential fistula formation. MRI particularly delineates intracranial pathology that complicates CSOM (Roland *et al.,* 1999) (Jung *et al.,* 2006).

Antibiotics are used to treat the infections but most of the organisms are acquiring resistance. In developing countries this problem is increasing rampantly due to misuse of antibiotics and inadequate antibiotic

treatment. Hence the aim of our study is to find out the fungal and bacterial aetiology and their antibiotic susceptibility pattern so as to know whether an empirical treatment based on the result of the study will effectively shorten the period of infection associated with discharge in patients of CSOM and thereby improve the surgical outcome.

Materials and Methods

This Descriptive cross sectional study was carried out in the Department of Microbiology of National Institute of Medical Sciences (NIMS) University, Jaipur, Rajasthan, India for a period of six months (February 2017 to July 2017). A total of 53 clinically diagnosed cases of CSOM attending ENT department of NIMS medical college was taken. Data regarding patient age, sex, duration of illness, previous medications and ears involved was obtained from the patient. The organisms isolated and identified provisionally as CSOM were identified further by using a standard protocol for identification (Koneman *et al.,* 2006).

Materials and Methods

Inclusion criteria

Adult and paediatric patients of both sexes and all age group who attend with the ear discharge of more than 2 weeks duration.

Exclusion criteria

Patient on systemic or topical antibiotics, patient with acute Suppurative otitis media i.e. duration of less than 2 weeks, Patients with cleft palate, Patients /parent /care giver who does not give his or her consent.

Isolation of various isolates was confirmed by several of test like colony characters, morphology (Gram's stain), pigment production, motility (by hanging drop) and other biochemical tests like Catalase, Oxidase test, Indole, Methyl red, Voges Proskauer, Citrate utilization test, Urease test and Triple sugar iron Test, Oxidation/fermentation for glucose, lactose, xylose, mannitol and maltose (Hugh and Leifson's media), Lysine and Ornithine decarboxylase and Arginine dihydrolase activity, ONPG test, Esculin test etc. were done for isolation of the gram negative and positive bacteria.

The following studies are necessary for isolation of fungi

Microscopic examination of sample by Gram staining and KOH Mount (Chandra, 2008)

Fungal culture

Sample was inoculated on SDA at 25^0C and 37^0C and was observed daily for fungal growth up to 4 weeks (Chandra, 2008)**.** The identification of species of fungal growth was done by various methods like Morphology of colony, Texture, Surface pigmentation, Microscopic examination like LPCB mount and slide culture (on chrome agar) were done to identify the fungi. Gram staining was done for identification of yeast and yeast like fungi, Chlamydospore formation and germ tube tests were done to identify *Candida albicans*, growth on corn meal agar.

Antimicrobial susceptibility

Antibiotic susceptibility testing was done by Modified Kirby Bauer's disc diffusion method as per CLSI guidelines 2017 using commercially available discs.

Results and Discussion

A total of 53 clinically diagnosed CSOM cases attending ENT department of NIMS medical college, Jaipur during the study

period were taken. In the present study, most of the CSOM cases i.e. 47 (89%) had unilateral ear involvement and only 6 (11%) had bilateral ear involvement. In patients with unilateral ear involvement most predominantly right ear i.e. 33(62%) patients were infected whereas left ear was involved in only 14 (27%) CSOM cases (Table 1 and 2).

In the current study, CSOM infection was found comparatively more in females patients 34 (64%) than in male patients 19 (36%). The male to female ratio was 1:1.8. Age group of 11-20 years (32%) and 21-30 years (21%) were predominantly affected (Table 3).

In the present study out of 53 ear swabs processed microbial growth was in 45 (85%) while 8 (15%) samples showed no growth (Table 4).

Out of 53 patients, 45 showed growth of bacteria and fungi. Pure growth (Monomicrobial) was obtained from 36 (68%) patients whereas 9 (17%) patients showed mixed growth (Polymicrobial) (Table 5).

Out of these 36 patients majority 32 (89%) showed pure bacterial growth only and 4 (11%) showed pure fungal growth (Table 6).

However, in remaining 9 (17%) patients mixed growth was obtained in which 2 (4%) patients showed mixed bacterial growth and 7 (13%) patients showed mixed bacterial and fungal growth. In the present study, most common organism isolated was *Pseudomonas species* (29%) then *Staphylococcus aureus* 20%. This was followed by CONS (12.50%), *Aspergillus niger* (9%), *Escherichia coli* (5%), *Candida albicans* (5%), *Proteus mirabilis* (3.57%), *Proteus vulgaris* (3.57%), *Klebsiella spp.* (3.57%), *Citrobacter* (1.79%), *Non fermentative* (1.79%), *Candida tropicalis* (1.79%), *Rhizopus* (1.79%) and *Alterneria* (1.79%) (Table 7).

In GPC most common organism isolated was *Staphylococcus aureus* 11(20%) followed by CONS 7(12.50%).

In GNB Most common organism isolated was *Escherichia coli* (5%) followed by *Proteus mirabilis* (3.57%), *Proteus vulgaris* (3.57%), *Klebsiella spp.* (3.57%), *Citrobacter* (1.79%).

In *Pseudomonas species* most common organism isolated was *Pseudomonas aeruginosa* 5(8.93%) followed by *Pseudomonas species* 11 (20%) and *Non-fermentative* 1(1.79%).

In fungal most common organism isolated was *Aspergillus niger* 5(8.93%) them *Candida albicans* 3 (5.36%) followed by *Candida tropicalis* (1.79%), *Rhizopus* (1.79%) and *Alterneria* (1.79%) (Fig. 1 and 2).

Table.1 Various comparative studies on side of ear involved in CSOM cases

Side involved	Present study (2017)	Ramesh Agarwal *et al.*, (2017)	Jeyakumari *et al.*, (2015)	Ruby Naz *et al.*, (2015)	Rakesh kumar *et al.*, (2013)
Unilateral	89 %	89.34 %	93%	73%	79%
Bilateral	11%	11	7%	27%	21%

Table.2 Various comparative studies on predominant side of infection in patients with unilateral ear involvement

Unilateral ear involvement	Present study (2017)	B.L. Choudhary *et al.*, (2014)	R Shyamala *et al.*, (2012)
Right ear	62%	60%	62%
Left ear	27%	34%	33%

Table.3 Various comparative studies on sex wise distributions of patients with CSOM infection

Gender	Present study (2017)	Ramesh Agarwal *et al.*, (2017)	Ruby Naz *et al.*, (2015)	Sushmita *et al.*, (2014)	Prakash M *et al.*, (2013)
Male	36%	58%	47%	58%	45%
Female	64%	42%	53%	42%	55%

Table.4 Various comparative studies on rate of isolation of microbial growth from CSOM cases

Isolation rate	Present study	Yadav Saurabh *et al.*, (2016)	Dhirendra kumar *et al.*, (2016)	Nawaz umar *et al.*, (2015)	Sunil kumar *et al.*, (2015)
Microbial growth	85 %	93 %	86 %	88.6 %	82 %
No growth	15%	7 %	14 %	14.4 %	18 %

Table.5 Various comparative studies on type of infection in CSOM cases

Infection	Present study	Yadav saurabh *et al.*, (2016)	Raakhee T *et al.*, (2014)	Kusuma Bai S *et al.*, (2013)	Prayaga N *et al.*, (2013)
Monomicrobial	68 %	85%	70%	66 %	39%
Polymicrobial	17 %	8%	15%	31 %	11%

Table.6 Various comparative studies on microorganism isolated from CSOM cases

Microorganism	Present study	Ramesh Agrawal *et al.*, (2017)	Asish J *et al.*, [88](2013)	Shrestha *et al.*,[126] (2011)
GNB	48%	46%	64%	51%
GPC	32%	47%	30%	38%
Fungus	20%	7%	6%	11%

Table.7 Comparative studies on various bacterial and fungal species isolated from CSOM cases

Organism	Present study	Geeta S.H. *et al.*, (2014)	Ramesh Agrawal *et al.*, (2017)	Dhirendra kumar *et al.*, (2016)	G jyoti Lakshmi *et al.*, (2014)	Ashish J *et al.*, (2013)
Pseudomonas spp.	28.57	17.92	41.1%	26%	41%	33%
Staphylococcus aureus	19.64	19.91	38.7%	38%	28.2%	25.8%
Coagulase Negative Staph. aureus	12.50	2.83	8.5%	-	2.5%	-
E.coli	5.36	1.26	2.3%	6%	5.1%	-
Citrobacter	1.79	6.28	1.5%	1.5%	2.5%	-
Proteus Mirabilis	3.57	9.12	1.5%	7%	-	20.6%
Proteus Vulgaris	3.57	1.57	3.1%	7%	7.6%	-
Klebsiella spp.	3.57	3.14	-	6%	3.8%	4.1%
Non fermenters	1.79	2.20	-	-	-	-
Candida albicans	5.36	1.57	2.3%	-	1.3%	1%
Aspergillus spp.	8.93	15.14	5.4%	-	3.8%	5.2%
Candida tropicalis	1.79	1.88	-	-	-	-
Rhizopus spp.	1.79	0.63	-	-	-	-
Alterneria	1.79	-	-	-	-	-

Table.8 Comparative studies on antibiotic sensitivity pattern of *Staphylococcus aureus* isolated from CSOM cases

Antibiotics	Present Study	Ramesh Agrawal *et al.*, (2017)	Dhirendra kumar *et al.*, (2016)	Jeyakumari *et al.*, (2015)	Rakesh kumar *et al.*, (2013)
Erythromycin	36	56	66.7	67	45
Clindamycin	73	94	85.2	93	70
Cotrimoxazole	55	-	83.3	33	-
Penicillin	9	26	-	7	-
Tetracycline	91	94	-	-	-
Linezolid	100	98	100	100	100
Vancomycin	100	90	100	100	100
Rifampicin	100	-	-	-	-
Ciprofloxacin	55	72	-	73	30
Gentamycin	91	-	-	-	-

Table.9 Comparative studies on MRSA prevalence in *S. aureus* isolates

	Present study	Ramesh Agrawal *et al.*, (2017)	Upasana Bhumbla *et al.*, (2016)	Dhirendra kumar *et al.*, (2016)	Bansal sulabh *et al.*, (2013)	Chakraborty *et al.*, (2013)
MRSA	27.27%	88%	35.7 %	7%	27.7 %	34.4 %

Table.10 Comparative studies on antibiotic sensitivity pattern of *Pseudomonas species* isolated from CSOM cases

Antibiotics	Present study	Ramesh Agrawal *et al.*, (2017)	Dhirendra kumar *et al.*, (2016)	Upasana Bhumbla *et al.*, (2016)	Ruby Naz *et al.*, (2015)
Cefepime	65	-	86.8		
Amikacin	65	83	71.1	66.7	75
Ticarcillin/Clavulanic Acid	94	-	-		-
Aztreonam	88	53	-		-
Ciprofloxacin	65	73	76.3	30.4	75
Ceftazidime	100	24	71.1	61.1	53
Meropenem	82	94	84.2	86.1	87
Gentamycin	88	-	73.7	27.3	81
Piperacillin	88	85	-		-
Piperacillin tazobactum	76	-	97.4	63.6	81
Colistin	88	-	-		93
Polymyxin- B	82	-	94.7		87

Table.11 Comparative studies on Antibiotic sensitivity pattern of *E. coli* isolated from CSOM cases

Antibiotics	Present study	Ramesh Agrawal *et al.,* (2017)	Jeyakumari *et al.,* (2015)	Rakesh kumar *et al.,* (2013)
Ceftazidime	33	33.3	40	-
Ceftazidime/Clavunic acid	67	66.	100	-
Meropenam	100	100	100	-
Gentamicin	67	66.6	-	77
Cefepime	33	-	-	-
Ampicillin	33	-	-	-
Amikacin	67	-	100	95.5
Piperacillin	67	-	-	76
Piperacillin/Tazobactum	100	-	100	90
Cefazolin	33	-	-	-

Table.12 antibiotic susceptibility pattern of various organisms in CSOM

Antibiotics	*Staph Aureus*	*CoNS*	*Citrobacter*	*E. coli*	*Proteus Vulgaris*	*Proteus mirabilis*	*Klebsiella spp*	*Pseudomonas aeruginosa*	*Pseudomonas spp.*	*Non-fermenters*
	%	%	%	%	%	%	%	%	%	%
Erythromycin	36	57	-	-	-	-	-	-	-	-
Clindamycin	73	57	-	-	-	-	-	-	-	-
Cotrimoxazole	55	14	-	-	-	-	-	-	-	-
Penicillin	9	14	-	-	-	-	-	-	-	-
Tetracycline	91	100	-	-	-	-	-	-	-	-
Linezolid	100	100	-	-	-	-	-	-	-	-
Vancomycin	100	100	-	-	-	-	-	-	-	-
Rifampicin	100	100	-	-	-	-	-	-	-	-
Ciprofloxacin	55	43	-	-	-	-	-	-	-	-
Gentamycin	91	71	100	67	100	100	50	60	100	100
Ceftazidime	-	-	0	33	50	100	50	100	100	100
Ceftazidime/Clavulanic acid	-	-	100	67	100	100	50	-	-	-
Meropenem	-	-	100	100	100	100	100	80	82	100
Cefepime	-	-	100	33	50	100	50	60	73	0
Ampicillin	-	-	100	33	50	100	50	-	-	-
Amikacin	-	-	100	67	100	100	100	60	64	100
Piperacillin	-	-	100	67	100	100	50	80	91	100
Piperacillin/Tazobactum	-	-	100	100	100	100	50	60	82	100
Cefazolin	-	-	100	33	100	50	50	-	-	-
Ticarcillin/Clavulanic Acid	-	-	-	-	-	-	-	100	91	100
Aztreonam	-	-	-	-	-	-	-	80	91	100
Ciprofloxacin	-	-	-	-	-	-	-	60	73	0
Colistin	-	-	-	-	-	-	-	100	82	100
Polymyxin- B	-	-	-	-	-	-	-	100	73	100

Fig. No. 1 : Microorganism Isolated from CSOM

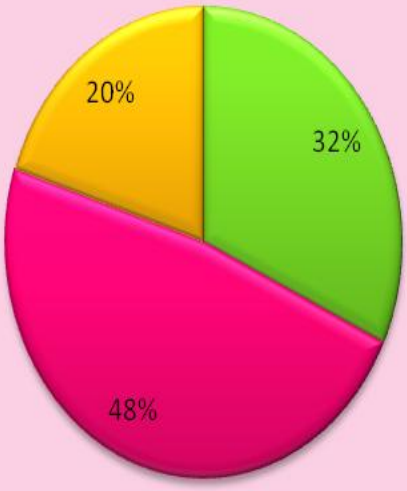


Fig. No. 2 : Various *species* isolated from CSOM

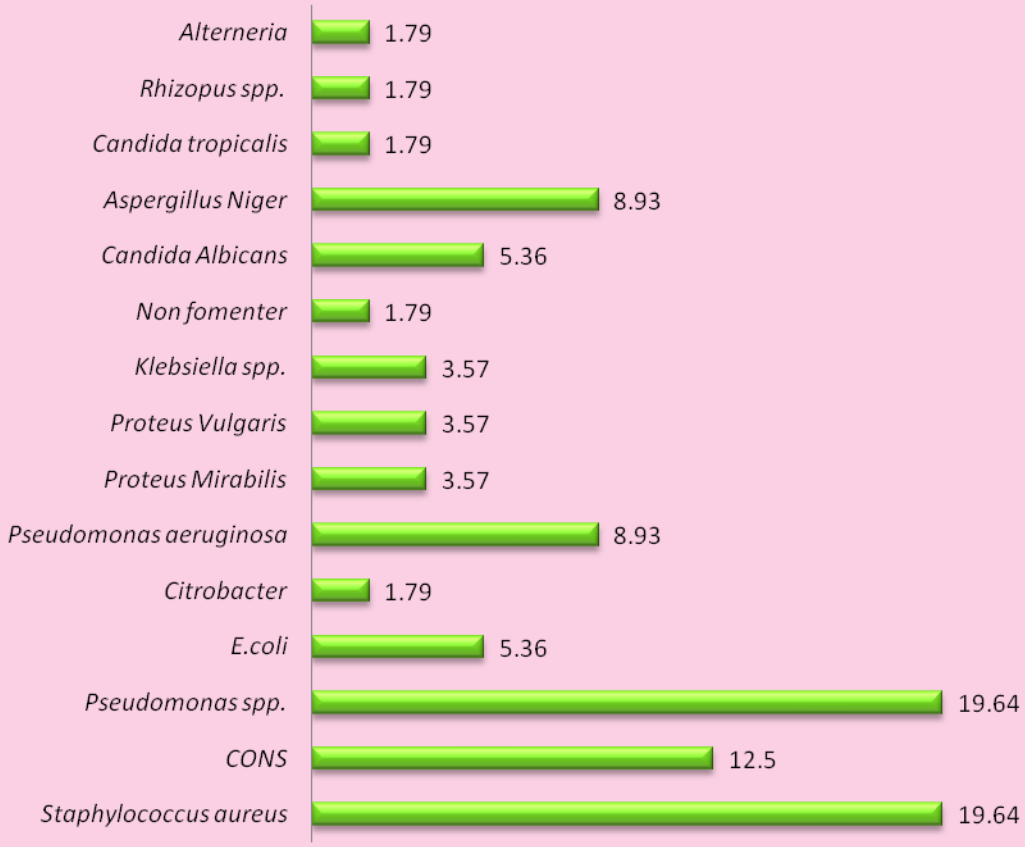
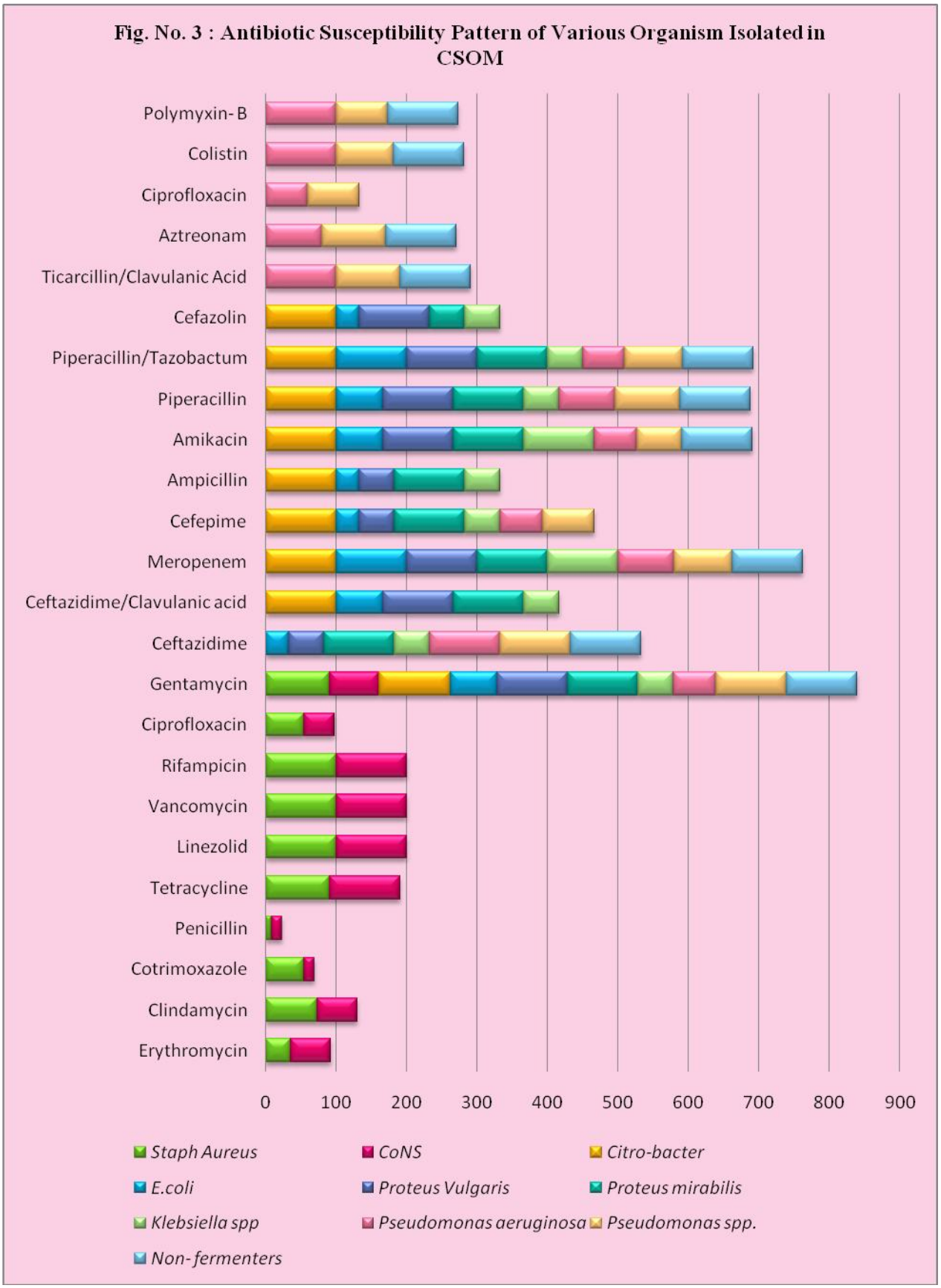


Fig. No. 3 : Antibiotic Susceptibility Pattern of Various Organism Isolated in CSOM



Antibiotic sensitivity testing results

Out of 11 *Staphylococcus aureus* isolates, all were sensitive to linezolid (100%), vancomycin (100%) and rifampicin (100%). *Staphylococcus aureus* showed decreased sensitivity to gentamycin (91%), tetracycline (91%), clindamycin (73%), cotrimoxazole (55%), ciprofloxacin (55%) and only erythromycin (36%) and penicillin (9%) sensitive.

Out of 7 CONS isolates all were sensitive to linezolid (100%), vancomycin (100%), rifampicin (100%) and tetracycline (100%). cons showed decreased sensitivity to gentamicin (71%), erythromycin (57%), clindamycin (57%), ciprofloxacin (43%) and only cotrimoxazole (14%) and penicillin (14%) sensitive.

In GNB, *Citrobacter* isolate was sensitive to all (100%) except ceftazidime (0%).

E. coli isolates, all were sensitive to meropenum (100%) followed by ceftazidime/clavulanic acid (67%), gentamycin (67%), amikacin (67%), piperacillin (67%), ceftazidime (33%), cefepime (33%), ampicillin (33%), cefazolin (33%) (Table 11).

Proteus vulgaris isolates, all were sensitive to ceftazidime/clavulanic acid(100%), meropenum (100%), gentamycin (100%), amikacin (100%), piperacillin(100%), piperacillin/tazobactum (100%) and cefazolin (100%) followed by ceftazidime (50%), cefepime (50%), ampicillin (50%).

Proteus mirabilis isolates were sensitive to all (100%) except cefazolin (50%).

Klebsiella species isolates, all were sensitive to meropenum (100%), amikacin (100%) followed by ceftazidime ceftazidime/clavunic acid (50%), gentamycin (50%), cefepime (50%), ampicillin (50%), piperacillin (50%), piperacillin/tazobactum (50%), cefazolin (50%).

In *Pseudomonas aeruginosa* isolates, all were sensitive to ticarcillin/clavulanic acid (100%), ceftazidime (100%), colistin (100%) and polymyxin-b (100%) followed by aztreonam (80%), meropenem (80%), piperacillin (80%), cefepime (60%), amikacin (60%), ciprofloxacin (60%), gentamycin (60%) and piperacillin/tazobactum (60%).

In *Pseudomonas species* isolates, all were sensitive to ceftazidime (100%) and gentamycin (100%) followed by ticarcillin/clavulanic acid (91%), aztreonam (91%), piperacillin (91%), meropenem (82%), piperacillin tazobactum (82%), colistin (82%), cefepime ciprofloxacin (73%), polymyxin-B (73%) and amikacin (64%).

In the present study, age group of 11-20 years (32%) were predominantly affected. This is similar to the study conducted by Rakesh Kumar *et al.,* (2013) showed the increased prevalence of CSOM in 30-40 years age in his study. In our study second most prevalent age group was 21-30 years and 31-40 years 21% each.

Antibiotic sensitivity and resistance of CSOM

In the present study, Out of 7 CONS isolates all were sensitive to linezolid (100%), vancomycin (100%) and rifampicin (100%) and tetracycline (100%). CONS showed decreased sensitivity to gentamicin (71%), erythromycin (57%), clindamycin (57%), ciprofloxacin (43%) and only cotrimoxazole (14%) and penicillin (14%) sensitive. which was correlated with the study of Ramesh Agrawal *et al.,* (2017) showed 100% sensitivity to linezolid, vancomycin (91%),

Clindamycin (91%), Penicillin (27%), Erythromycin (46%) and Ciprofloxacin sensitive to 73% strains. A study conducted by Hirapure *et al.*, (2014) showed 71% sensitivity to gentamicin and cotrimoxazole sensitive to 58% cases (Table 12 and Fig. 3).

In the present study out of 2 *Proteus mirabilis* isolated, all were sensitive to all (100%) except cefazolin (50%). *Proteus mirabilis* was sensitive to gentamicin (95%), chloramphenicol (90%), tobramycin (90%), cefotaxime (90%), ceftriaxone (90%), piperacillin tazobactum (85%), and gatifloxacin (85%). 100% *Proteus mirabilis* isolated showed sensitivity to ceftazidime and ciprofloxacin. This was seen in study done by Mandana *et al.*, (2011) and Osazuva *et al.*, (2011) with similar sensitivity to Gentamicin.

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