



A CLINICAL, BACTERIOLOGICAL, MYCOLOGICAL PROFILE AMONG PATIENTS WITH CHRONIC SUPPURATIVE OTITIS MEDIA IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Background and Objectives: Chronic suppurative otitis media (CSOM) is defined as a perforation of the tympanic membrane, which has been prevented from healing by chronic inflammation and has persisted for more than 3 months, with a history of discharge for atleast 6 weeks. It is the most commonly encountered disease in otolaryngology practice. Due to advent of newer and sophisticated antibiotics, microbial flora is changing constantly. Culture of the middle ear fluid constitute a valuable tool for definitive diagnosis, to guide therapy, to evaluate treatment failures and for research studies to determine the efficacy of antimicrobials against the most common agents. Many studies have been done on aerobic bacteria and fungal isolates causing CSOM but very few studies are done on anaerobic bacteria causing CSOM. Hence, the present study will be undertaken to know the microbiological profile of CSOM including anaerobes.

Objectives of the study:

- (1) To estimate the prevalence of bacterial (aerobic and anaerobic) and mycotic flora in middle ear infection among clinically diagnosed chronic suppurative otitis media patients.
- (2) To assess the antibiotic sensitivity pattern of the aerobic bacterial isolates.

Methods:

Hundred clinically diagnosed cases of CSOM of all age groups and both the sexes attending the ENT OPD with either unilateral or bilateral ear discharge were considered in this study. Following a detailed questionnaire, four ear swabs were taken from each patients and processed in Microbiology laboratory. Aerobic and anaerobic bacterial and fungal isolates were identified and corresponding antimicrobial sensitivity tests were done for aerobic bacterial isolates based on standard methods. **Results :** Among the aerobic bacterial isolates *Pseudomonas aeruginosa* (40%) was the most predominant bacteria isolated followed by *Staphylococcus aureus* (21%). The most common anaerobic bacteria was *Finigoldia magna* (25%) followed by *Fusobacterium necrophorum* (18.75%). *Aspergillus niger* (35%) was the most predominant fungus followed by *Candida albicans* (25%). Gram positive cocci were highly sensitive to Vancomycin, Teicoplanin, Tetracycline. Gram negative bacilli belongs to Enterobacteriaceae family were highly sensitive to Ciprofloxacin and Cefepime. Gram negative non fermenters were highly sensitive to Amikacin, Ticarcillin Clavulanic acid. **Conclusion:** Due to emergence of multidrug resistant organisms appropriate antibiotics should be selected before starting treatment.

KEYWORDS

CSOM, Aerobic, anaerobic isolates and fungal isolates, *Pseudomonas aeruginosa*, *Finigoldia magna*, *Aspergillus niger*, Vancomycin, Amikacin

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INTRODUCTION

Ear is an important sensory organ. It is worthy to note that ear infections are a very common problem worldwide. Ear infection is an inflammation of the ear and ear discharge is one of the commonest symptoms of ear infection. Ear infection occur in all age groups. Ear discharge may arise from external auditory meatus in otitis externa or middle ear cavity in otitis media.²

Otitis media is an inflammation of the middle ear caused by bacteria, fungi and virus resulting in inflammation of the mucosal lining. It can be acute, subacute and chronic. The acute form is Acute Suppurative Otitis Media (ASOM) usually associated with the infection in the upper respiratory tract whereas persistent form is known as chronic suppurative otitis media (CSOM).³

Chronic suppurative otitis media (CSOM) is defined as a perforation of the tympanic membrane, which has been prevented from healing by chronic inflammation and has persisted for more than 3 months, with a history of discharge for atleast 6 weeks.⁴

The disease is classified in to two types depending upon whether the disease process affects the pars tensa or the pars flaccid of the tympanic membrane.⁵

- (1) Tubotympanic
- (2) Attico-antral

Tubotympanic is known as the safe type or benign type as there are no serious complications whereas, attico-antral is known as the unsafe or dangerous type because of its associated complications and at times, it

may be life threatening.⁶

The incidence of chronic suppurative otitis media is higher in developing countries, especially amongst the lower and the middle socio-economic status of the society (with an urban: rural ratio of 1:2) because of poor nutrition, improper hygiene and lack of health education.⁷

It usually begins as a complication of persistent acute otitis media with perforation in childhood. It affects almost fifty percent of the children by the age of 5 years. CSOM has direct impact on the hearing of patient causing conductive and sensorineural hearing loss and also on child development (WHO, 1998)⁸

The worldwide prevalence of CSOM is 65-330 million people; and 39-200 million (60%) suffer from clinically significant hearing impairment. India is one of the countries with highest CSOM prevalence (>6%) where urgent attention is needed.⁹

Infection is usually bacterial (aerobic and anaerobic) in origin; however, viral and fungal agents have also been isolated.¹⁰ Various studies have shown that both gram positive and gram negative aerobic and anaerobic organisms are responsible for the infections of the middle ear, gram negative organisms outnumbering the gram positive ones.

The most common aerobic causative isolates are *Staphylococcus aureus* and *Pseudomonas aeruginosa*. Others include *Proteus* species, *E. coli*, *Klebsiella* species, *Streptococcus* species, *H influenzae*, Gram negative non fermenting bacilli.¹¹

The most common anaerobic causative isolates are Bacteroides species, Peptostreptococcus species, Fusobacterium species, and Propionibacterium.¹²

Aspergillus species and Candida species are most fungal isolates which are responsible for CSOM.¹¹

The treatment of CSOM is controversial and subject to change particularly in the developing countries, the prevalence and antibiogram of these organisms has been reported to vary with time and geographical area as well as continent to continent, probably due to indiscriminate use of the antibiotics. Hence, the periodic update of prevalence and antibiogram of the etiological agents for CSOM would be helpful in therapy and management of patients.¹³

Therefore the present study was undertaken to know the new trend of prevalence of CSOM, different bacteria (aerobic and anaerobic) and fungi causing CSOM and antibiogram of aerobic bacterial agents in CSOM among the people.

MATERIALS AND METHODS

Source of data:

The present study "A Clinical, Bacteriological and Mycological Profile Among Patients With Chronic Suppurative Otitis Media in a Tertiary Care Hospital" was carried out in the Department of Microbiology, Vydehi Institute Of Medical Science And Research Centre, Bangalore, Karnataka on patients of any age and sex diagnosed as suffering from CSOM after thorough clinical evaluation by an ENT surgeon attending ENT OPD.

Study design:

Cross sectional study

• Study period:

The study was conducted for 18 months (January 2017 to June 2018)

Study setting:

Clinically diagnosed CSOM patients attending in ENT OPD, Vydehi Institute Of Medical Science and Research Centre.

Inclusion criteria:

i) All clinically diagnosed CSOM male and female patients of all age group presenting to the OPD with purulent ear discharge (unilateral/bilateral) for more than 3 months, attending the ENT Department of Vydehi Institute Of Medical Science & Research Centre were considered.

ii) Patients who are not on any antibiotic treatment last 48 hours.

Exclusion criteria:

I) Patients who are already on systemic and topical antibiotic treatment of CSOM.

ii) Patients on follow up in the OPD.

Sampling technique:

All the clinical samples (ear swabs) were collected from ENT OPD and the study (culture and sensitivity) was conducted in the Department Of Microbiology.

History taking and examination:

A proforma was filled for each patient documenting age, sex, socio economic status and clinical information, including chief complaints, duration of symptoms were noted.

Sample collection:

Ear discharge was collected under strict aseptic precautions from the affected ear of CSOM patients. Excess discharge was mopped up from external auditory canal and it was cleaned with 70% alcohol first and was allowed to dry for 30-40 seconds to achieve sterile area.²⁵ Then with four sterile swabs specimens were collected. One swab was for gram staining and wet mount preparation using 10% KOH. The second swab was subjected for aerobic culture on Blood agar, MacConkey agar and Chocolate agar. The third one was used for anaerobic culture by inoculating in Robertson Cooked Meat media soon after the collection of the specimen. The fourth swab was used for mycological culture on two Sabouraud's dextrose agar plate

without antibiotics.^{76,77}

Sample processing:

Day 1.

Direct microscopy:

With the first swab a smear was made on a clean grease free glass slide and was fixed by heating. Gram's staining was done for the smears so made and observed under oil immersion. The smear was screened carefully for the presence of bacteria, their number and their gram reaction (gram positive/ negative), shape, size and arrangement and also for the presence or absence of pus cells.

Wet mount preparation using 10% KOH was done by using the same swab for the fungal elements.

For aerobic culture

The second swab was inoculated on a dried plates of 5% sheep Blood agar, MacConkey agar, and Chocolate agar by rolling the swab over the agar to make a primary well and then streaking from the primary inoculation using a sterile bacteriological loop to form secondary, tertiary and quaternary streak lines. These plates were incubated aerobically at 37°C for 24 hours and looked for evidence of growth For anaerobic culture:

The third swab which was immediately inoculated in RCM broth after sample collection, incubated for 48 hours.

For fungal culture:

The fourth swab was inoculated on two Sabouraud's dextrose agar without antibiotics. One plate was incubated at 25°C and another at 37°C.

Day 2

• Aerobic culture plates were observed for growth. If growth was present,

i) Colony characteristics were observed

ii) Smears were prepared from the colony and gram staining was performed

iii) Biochemical tests were done according to the procedures described in Mackie and MacCartney Practical Microbiology and Koneman's Text Book Of Diagnostic Microbiology

iv) Antibiotic susceptibility testing of the isolates was done on Mueller Hinton agar using Kirby Bauer disc diffusion method.

Method: Bacterial suspension was prepared by inoculating few isolated colonies of similar morphology in to 4-5 ml of peptone water and incubated at 37°C for 2-4 hours. The turbidity of the growth was adjusted to 0.5 MacFarland turbidity standards and lawn culture was made on the surface of the medium with sterile cotton swab. The selected antibiotic discs were then placed aseptically on this media 1.5 cm from the edge of the plate and 2.5 cm apart from each other using a sterile forceps. The plates were then incubated at 37°C for 18-24 hours. The zone of inhibition were measured and reported as sensitive, intermediate or resistant. Commercially available Himedia discs were used.

• If no growth was obtained, the culture plates were incubated further for 24 hours.

SDA plates were examined for growth. If growth was present

i) Colony characteristics were observed

ii) Gram smear was prepared for yeast like fungus and germ tube test was done for species identification

iii) For mycelia colonies LPCB tease mount was prepared to observe its characteristics. The isolates were subjected to slide culture.

If no growth observed, SDA plates were incubated further for 1 week.

Day 3

For aerobic culture:

The identification tests and antibiotic sensitivity tests were interpreted and reported.

Anaerobic culture:14

Smear was made on a grease free clean glass slide, gram stain was done followed by methanol fixation and subcultures done on to two 5% sheep Blood agar and one Neomycin Blood agar and one Chocolate agar plate along with 5U metronidazole disc for presumptive identification of anaerobes. One 5% Sheep Blood agar and one Neomycin Blood agar were incubated anaerobically using McIntosh Fildes jar and Gas pack for 3 to 4 days. Methylene blue tablet was used as indicator. Another 5% Sheep Blood agar and Chocolate agar plate was incubated in CO₂ incubator for aerotolerance test.

Metronidazole sensitive colonies were subjected for gram stain. Identification of bacterial isolates:15

(A) Aerobic bacteria:

The bacterial isolates were identified based on the biochemical reactions, as described in standard microbiological textbooks- Mackie and Mc Cartney and Konemann.

(B) Anaerobic bacteria:

Presumptive Identification of anaerobic bacteria was done depending on colony morphology and gram staining.

Chart 7: PRESUMPTIVE IDENTIFICATION OF ANAEROBIC BACTERIA BASED ON COLONY MORPHOLOGY AND GRAM STAIN14

ANAEROBES	COLONY MORPHOLOGY	GRAM STAIN
Bacteroides spp	Neomycin Blood agar- Round, opaque, grey white colonies	Pleomorphic Gram negative rods, some with vacuoles giving safety pin appearance
Prevotella spp	Neomycin Blood agar- small, opaque, grey colonies which fluoresce red under UV light and colonies become black on prolonged incubation	Small, Gram negative coccobacilli
Fusobacterium spp	Neomycin Blood agar- small, moist, opaque grey white to translucent colonies	Fusobacterium necrophorum- long gram negative bacilli with rounded end Fusobacterium nucleatum- pale staining gram negative spindle (pointed and tapered ends) shaped rods
Finegoldia magna	Neomycin Blood agar- small semi transparent, grey coloured beta hemolytic colonies	Large gram positive cocci in tetrads or cluster (similar in size and appearance to staphylococcus)
Peptostreptococcus anaerobius	Neomycin Blood agar- opaque grey coloured colonies	Gram positive cocci arranged in small cluster and chains
Anaerococcus spp.	Neomycin Blood agar- small, translucent in grey coloured colonies	Gram positive cocci arranged singly, in pairs and rarely in short chains
Actinomycetes spp.	Neomycin Blood agar- smooth, glistening transparent colonies with an entire or furred edge	Gram positive rod shaped filamentous bacilli (ray like appearance)

Identification of the anaerobic isolates at species level was carried out by Automated VITEK 2 compact system (Biomérieux) using VITEK ANC ID cards.

The ANC card is based on established biochemical methods and newly developed substrates. There are 36 biochemical tests measuring carbon source utilization and enzymatic activities. final results are available in approximately six hours.

Procedure: The inoculum was prepared from a pure culture, according to good laboratory practices. Isolated colonies were selected from a primary plate if culture requirement were met or subculture of the

organism to be tested was done to Neomycin Blood agar plate and incubated anaerobically. Morphologically similar colonies were transferred to the saline tube (which contain 3.0 ml of sterile saline) and a homogenous organism suspension with a density equivalent to a McFarland No. 2.70 to 3.30 using a calibrated VITEK 2 DensiCheck Plus. Place the suspension tube and ANC card in the cassette. Refer to the appropriate instrument user manual for the instructions of data entry and how to load the cassette in to the instrument. In addition to the internal tests included on the card, three offline tests are required in the ANC ID algorithm. The offline tests selected for use in the ANC ID product are gram stain, morphology, and aerotolerance. The ANC offline test results can be entered at the Smart Carrier Station or the workstation.

VITEK 2 system identify the organism by using a methodology based on the characteristics of the data and knowledge about the organism and reactions being analyzed.

ANC Quality control: Clostridium septicum ATCC 12464

Bacteroides ovatus ATCC BAA- 1296

Identification of fungal isolates:

(a) Identification of yeast and yeast like fungus:

Gram staining was done for identification of budding yeasts and yeast like fungi and Germ tube test was done to differentiate between Candida albicans and Candida non albicans.

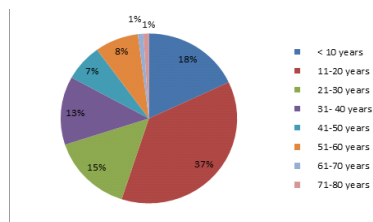
(b) Identification of moulds:

LPCB tease mount and slide culture were done to identify the filamentous fungi. Repeat sample was collected after 1 week to rule out laboratory contaminants.

RESULTS

The present study "A clinical, bacteriological and mycological profile among patients with chronic suppurative otitis media in a tertiary care hospital" was conducted over a period of 18 months from January 2017 to June 2018 at Vydehi Institute of Medical Sciences and Research Centre, Bangalore, Karnataka. A total of 100 clinically diagnosed cases of Chronic suppurative otitis media attending ENT OPD were studied.

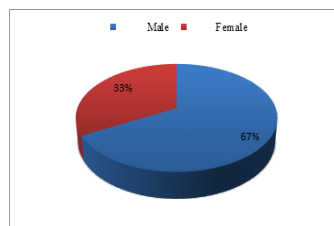
(1) Age ,sex and socioeconomic status distribution of CSOM cases:



Graph 1: Age distribution of CSOM cases

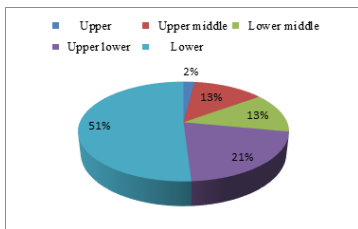
Age range was from 1 year to 80 years. Out of 100 patients maximum number of patients fell in the age group 11-20 years (37%), followed by 1-10 years (18%) ; 21-30 years (15%); 31-40 years (13%); 51-60 years (8%) and 41-50 years (7%) age group. Least number of cases fell in the age group 61 to 70 years (1%) and 71-80 years (1%).

Mean age for all 100 patients was 24.9 and standard deviation was 15.8462



Graph 2: Sex distribution of CSOM cases

Out of 100 clinically diagnosed CSOM patients 67 (67%) was male and 33 (33%) was female. Male : female ratio was 2:1.



Graph 3: Socio economic distribution of CSOM cases

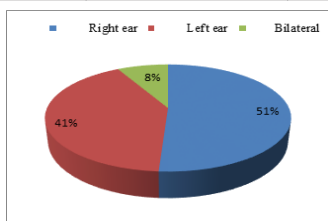
Out of 100 patients majority (51%) was belong to lower socio economic class followed by upper lower(21%), lower middle (13%) and upper middle(13%).Least number of cases (2%) were belong to upper class.

Socio economic scale was calculated according to Modified Kuppuswamy socioeconomic scale⁹⁷

(1) Distribution of CSOM cases according to the site of involvement:

Table 4: Distribution of CSOM cases according to the site of involvement

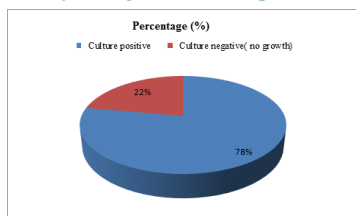
Site of involvement	Frequency	Percentage (%)
Right ear	51	51
Left ear	41	41
Bilateral	8	8
Total	100	100



eGraph 4: Distribution of CSOM cases according to the site of involvement

Out of 100 cases were found to have unilateral involvement with right ear (51%) being more commonly affected as compared to the left ear (41%). 8 cases (8%) were found with bilateral involvement. Total 108 samples were collected from 100 cases. Two samples each were collected from bilateral cases.

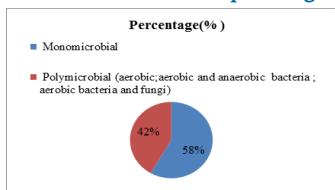
(3) Culture positivity among the CSOM samples:



Graph 5: Culture positivity among the CSOM samples

Among the 108 samples were collected from 100 clinically diagnosed CSOM cases 84 (77.77%) samples were culture positive showing either aerobic or aerobic and anaerobic or aerobic and fungal or fungal growth, whereas 24 (22.22%) samples were culture negative showing no growth

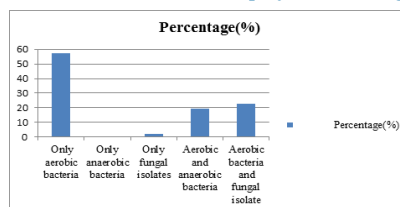
(4) Organism wise distribution of culture positive growth:



Graph 6: Organism wise distribution of culture positive growth

Out of 84 culture positive samples, 49 samples (58.33%) samples were positive for monomicrobial growth and polymicrobial (aerobic or aerobic and anaerobic bacteria or aerobic bacteria and fungus) growth were isolated from 35 samples (41.66%) .

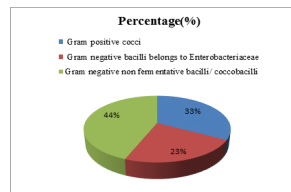
(5) Distribution of monomicrobial and polymicrobial organisms:



Graph no 7: Distribution of monomicrobial and polymicrobial organisms:

Among 84 culture positive samples, 83(98.80%) were positive for aerobic bacteria, and 1(1.19%) sample was positive for fungal isolate. Out of 83 aerobic culture positive samples, only aerobic bacteria were isolated from 48 (57.14%) samples; aerobic and anaerobic bacteria were isolated from 16 (19.05%) samples; aerobic bacteria and fungal isolates were isolated from 19(22.62%) samples.

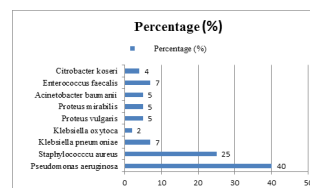
(6) Distribution of aetiological agents in culture positive cases: Pattern of aerobic bacteria:



Graph no 8: Pattern of aerobic bacteria:

Out of 83 aerobic culture positive samples, 27 (33%) were gram positive cocci, gram negative bacilli belongs to Enterobacteriaceae family were isolated from 19 (23%) samples and gram negative non fermentative bacilli / coccobacilli were isolated from 37(44%) samples.

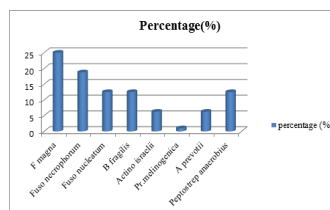
Distribution of aerobic bacterial isolates in culture positive samples



Graph 9: Distribution of aerobic bacterial isolates in culture positive samples :

Out of 84 culture positive samples, a total number of 10 different aerobic bacteria were isolated from 83 (98.80%) samples. The most common organism was Pseudomonas aeruginosa (40%) followed by Staphylococcus aureus (25%), Klebsiella pneumoniae (7%), Enterococcus faecalis(7%), Proteus mirabilis (5%), Proteus vulgaris (5%), Acinetobacter baumannii(5%), Citrobacter koseri (4%) and Klebsiella oxytoca(2%).

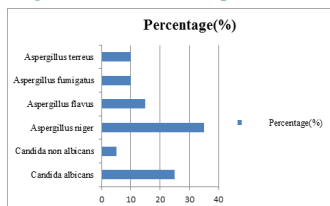
Distribution of anaerobic bacterial isolates in culture positive samples:



Graph 10: Distribution of anaerobic bacterial isolates in culture positive samples

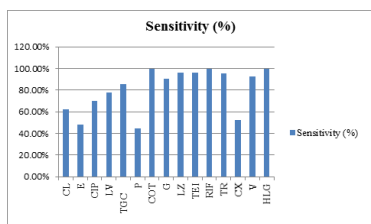
Out of 84 culture positive samples a total number of 8 different types of anaerobic organisms were isolated from 33(39%) samples. *Finegoldia magna* (25%) was the most common organism followed by *Fusobacterium necrophorum* (18.75%), *Bacteroides fragilis*(12.5%), *Fusobacterium nucleatum*(12.5%), *Peptostreptococcus anaerobius* (12.5%), *Actinomyces israelii* (6.25%), *Prevotella melanogenica*(6.25%) and *Anaerococcus prevotii* (6.25%).

Distribution of fungal isolates in culture positive samples



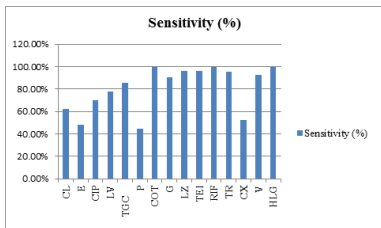
Graph 11: Distribution of fungal isolates in culture positive cases:

Out of 84 culture positive samples a total 6 different types of fungal isolates were isolated from 20 samples. *Aspergillus niger* (35%) was the most common fungal isolate followed by *Candida albicans*(25%), *Aspergillus flavus* (15%), *Aspergillus fumigatus* (10%), *Aspergillus terreus*(10%) and *Candida non albicans*(5%)



(7) Antibiotic susceptibility pattern:

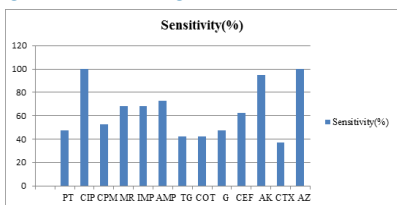
(i) Gram positive cocci:



Graph 12: Antibiotic susceptibility of Gram positive cocci:

Among 21 *Staphylococcus* isolates all 21 strains were sensitive to Linezolid (100%), Teicoplanin(100%), Rifampicin (100%) and Vancomycin(100%) [according to CLSI guidelines MIC of Vancomycin was tested by VITEK2. Based on VITEK2 database, the mean MIC of vancomycin was $\leq 0.5 \mu\text{g/ml}$] followed by Tetracycline (94.44%) Ciprofloxacin(88.88%), Gentamycin(88.88%) and Tigecycline(83.33%) *Enterococci faecalis* were highly sensitive to High level Gentamycin(100%) followed by Linezolid(83.33%), Teicoplanin(83.33%) and Vancomycin(66.66%)

(ii) Gram negative bacilli belongs to Enterobacteriaceae



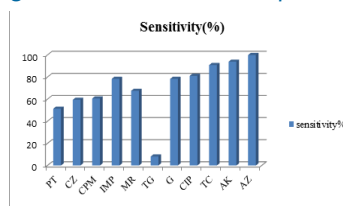
Klebsiella pneumoniae strains were highly sensitive to Ciprofloxacin(100%), Cefepime(100%) followed by Cotrimoxazole (83.33%) and Amikacin (83.33%) whereas *Klebsiella oxytoca* isolates

were highly sensitive to Cotrimoxazole(100%), Ciprofloxacin (100%) followed by Imipenem(50%), Meropenem(50%)

Proteus vulgaris isolates were highly sensitive to Ciprofloxacin(100%), Ampicillin(100%),Amikacin(100%) followed by Meropenem(75%), Imipenem(75%), whereas *Proteus mirabilis* were highly sensitive to Ciprofloxacin(100%) and Amikacin(100%).

Citrobacter koseri strains were highly sensitive to Piperacillin/tazobactam(100%), Ciprofloxacin(100%), Imipenem (100%) followed by Tigecycline(66.6%)

(iii) Gram negative non fermentative bacilli/ coccobacilli:



Chronic suppurative otitis media is one of the most common clinical ear diseases which is encountered in clinical practices. Because of variation in patient community and due to inadvertent use of antibiotics, the pattern of microbiological flora varies in chronic suppurative otitis media. Early bacteriological diagnosis of all cases will assure accurate and appropriate effective therapy. Treatment hence needs to be instituted early and effectively to avoid complications. In the present study an attempt has been made to identify the aerobic and anaerobic bacterial and fungal profile of CSOM patients attending the ENT Department of Vydehi Institute Of Medical Sciences and Research Centre and also to determine the antibiotic sensitivity pattern of the aerobic bacterial isolates. Distribution of CSOM cases with respect to age, sex and socioeconomic status, site of ear involvement were also studied. The results are compared with the other studies and described as follows:

Age distribution:

In the present study maximum number of patients were between the age group 11-20 years(37%) followed by the age group under 10 years of age(18%); so more than half of the patient (55%) were less than 20 years of age. The incidence of CSOM decreased with the increase in age. This was in accordance with the study conducted by Y. K. Harshika et al(2015)16, R.Prakash et al(2015)17, Shazia Parveen and Janardhan Rao(2012)18 whereas in a study done by Mary Nirmala S et al (2016)19, the most prevalent age group was 41 to 50 years and Moorthy et al(2013)20 found that there was no age predisposition. Higher incidence of otitis media in first two decades may be due to

- (i) Abundance of lymphoid tissue in children may obstruct the Eustachian tube
- (ii) Increased risk of respiratory infection.
- (iii) Decreased immunocompetence
- (iv) Short and straight Eustachian tube in infants and young children allows ready access of bacteria to middle ear.21,22

Sex distribution:

In this study, there was male predominance as compared to females. The sex ratio male: female was 2:1. Similar findings were obtained by Sangodan Rajesh and Nirmala Sukumar (2017),21Y.K.Harshika et al(2015)16,Saranya SK et al(2015)3. But Jial Deepak et al(2013)6, Raakhee T et al(2014)2, Prakash M et al(2013)22 showed more incidence of CSOM in females as compared to males.

Male predominance may be due to their more exposed way of life style. Socio economic distribution:

The present study revealed high incidence of CSOM cases seen in lower socio economic class(51%) followed by upper lower class(21%). Similar finding was obtained by Geeta S.H (2014) 23 where as Sachin Garud et al(2016) 24 have reported the prevalence of CSOM was more common in upper lower class(37.5%) followed by lower class(34.37%)

Incidence of CSOM observed in lower socio economic group could be attributed to many factors such as overcrowding, poor nutrition,

unhygienic conditions, improper and inadequate medical care, illiteracy and lack of health consciousness.

Distribution of CSOM according to the site of involvement:

In the present study unilateral [right ear(51%)> left ear(41%)] ear infection was more common than bilateral (8%). Similar findings were obtained in the study conducted by R Shyamala et al(2012)²⁵, Sahira Haneefa et al(2017)²⁶

Microbial profile:

In the present study, out of 108 samples 84(77.77%) were culture positive among which 49(58.33%) samples were positive for monomicrobial growth and 35(41.66%) samples were positive for polymicrobial growth (either aerobic bacteria or aerobic bacteria and anaerobic bacteria or aerobic bacteria and fungi), whereas 24(22.22%) samples showed no growth. Among 84 culture positive samples, 27(33%) were gram positive cocci and 19(23%) were gram negative bacilli belongs to Enterobacteriaceae family and 37 were (44%) gram negative non fermentative bacilli/ coccobacilli. The predominant bacteria isolated was *Pseudomonas aeruginosa*(40%), followed by *Staphylococcus aureus*(25%), *Klebsiella pneumoniae* (7%). These findings were similar to the study done by Abida Khatun et al(2015)²⁷, Sreeshma Balan et al(2017)²⁸ whereas Wadile Rahul Gopichand et al (2015)²⁹, Sowmya Tumkur et al(2017)³⁰ showed that *Staphylococcus* was more predominant organism followed by *Pseudomonas aeruginosa*.

The occurrence of *Pseudomonas aeruginosa* as prime offender can be attributed to:

(a) *Pseudomonas* survives competition with other pathogens due to minimal nutritional requirement and its armamentarium of antibacterial products like pyocyanin and bacteriocin.

(b) Vartiainen Eero and Vartiainen Jukka postulated *Pseudomonas* has ability to carve a niche for itself in local infection through necrotizing activities of its extracellular enzymes.

(c) In addition, the organism acts as an opportunistic pathogen, flourishes in the external auditory canal and may cause supportive disease in contiguous sites.

The role of anaerobes in CSOM is often questioned as they are mostly detected in cases with extensive cholesteatoma or granulation tissue however, it is advocated that while investigating pathogenic organisms in CSOM requests for anaerobic culture should be included and the medical therapy should be directed at the eradication of the pathogenic aerobic and anaerobic organisms.⁶⁴ In the present study out of 84 culture positive samples anaerobic isolates were yielded from 33(39%) samples. *Finegoldia magna* /*Peptostreptococcus magnus* (25%) was most predominant bacteria, followed by *Fusobacterium* spp[*Fusobacterium necrophorum*(18.75%) and *Fusobacterium nucleatum*(12.5%)], *Bacteroides fragilis*(12.5%), *Peptostreptococcus anaerobius* (12.5%). This finding was similar to the study done by Geeta S (2014)²³ where *Peptostreptococcus* spp(45.6%) were most predominant bacteria whereas O Satyanarayan et al (2015)³¹, Rajat Prakash et al(2013) have reported *Bacteroides fragilis* and *Clostridium* spp as predominant bacteria followed by *Peptostreptococcus* spp and *Fusobacterium* spp.

In the present study, fungal etiology found in 20 (23.80%) samples out of which *Aspergillus niger* had the highest prevalence of 35% followed by *Candida albicans*(25%). Similar findings were obtained by Geeta S(2014)²³, Chandra Bhan et al(2017)³¹ and Sreeshma Balan et al (2017)²⁸ where *Aspergillus niger* was the predominant fungus followed by *Candida* spp. However, a study done by Harvinder Kumar et al(2011) ⁷ reported *Candida albicans*(60%) as predominant fungus followed by *Aspergillus fumigatus* (20%).

Antibiogram:

In the present study among the all aerobic isolates *Pseudomonas aeruginosa* was the predominant bacteria followed by *Staphylococcus aureus* and *Klebsiella* spp. *Pseudomonas aeruginosa* isolates were most sensitive to Amikacin(93.93%) followed by Ticarcillin Clavulanic acid (90.90%), Ciprofloxacin (87.87%) and Gentamycin(81.81%). Similar findings were obtained by Prakash M et al(2013)²², Y. K.

Harshika et al(2015)¹⁶ and V Rama Chandra Rao et al(2014) where they showed that *Pseudomonas* isolates were highly sensitive to Amikacin followed by Ciprofloxacin and Gentamycin. However a study done by Nagraj M et al(2018)³² showed *Pseudomonas* isolates were most sensitive to Piperacillin Tazobactam.

In the present study it was found that the second most common bacteria *Staphylococcus aureus* isolates were 100% sensitive to Vancomycin, Linezolid, Cotrimoxazole and highly sensitive to Tetracycline(95.23%) followed by Gentamycin (90.47%) and Ciprofloxacin(85.71%). These findings were similar to the study done by Arvind N et al(2014)³³, Y.K. Harshika et al(2015)¹⁶, Sowmya Tumkur et al(2017)³⁰ and Nagraj M et al(2018)³² where *Staphylococcus aureus* isolates were 100% sensitive to Vancomycin and Linezolid and highly sensitive to Gentamycin and Cotrimoxazole. However a study done by Mohammed Jamu Kazeem et (2016)³⁴ in Northern Nigeria showed that *staphylococcus aureus* isolates showed a sensitivity of 91.9% to Levofloxacin followed by Gentamycin(88.4%).

The third most common bacteria *Klebsiella* species found in the present study were 100% sensitive to Ciprofloxacin and Cefepime followed by Cotrimoxazole(83.33%) and Amikacin(83.33%). Similar findings were obtained by Raakhee T et al(2014)² whereas a study done by V.Rama Chandra et al(2014)³⁵ showed *Klebsiella* isolates were 100% sensitive to Gentamycin and Ceftriaxone followed by Amikacin(93.7%)

SUMMARY

The present study “ A clinical, bacteriological and mycological profile among patients with chronic suppurative otitis media in a tertiary care hospital” was carried out in the Department of Microbiology of Vydehi Institute Of Medical Science and Research Centre, Bangalore on 100 clinically diagnosed CSOM patients attending ENT OPD were studied for bacteriological (aerobic and anaerobic) and fungal flora from January 2017 to June 2018.

Ear discharge was collected under strict aseptic precautions from the affected ear of CSOM cases. Excess discharge was mopped up from external auditory canal and it was cleaned with 70% alcohol first and was allowed to dry for 30-40 seconds to achieve sterile area. Then with four sterile swab specimens were collected. One swab was subjected for direct microscopy and wet mount preparation using 10% KOH. The second swab was subjected for aerobic culture on Blood agar, MacConkey agar and Chocolate agar. The third one was used for anaerobic culture by inoculated in Robertson Cooked Meat media soon after the collection of the specimen. The fourth swab was used for mycological culture on two Sabouraud's dextrose agar plate without antibiotics. After isolation, Gram stain, biochemical reactions were done for bacterial isolates and LPCB mount, slide culture were done for fungal isolates according to the standard procedures. Identification of the anaerobic bacterial isolates was done by using VITEK 2 ANC cards. Antibiotic susceptibility testing of the aerobic bacterial isolates was done on Mueller Hinton agar using Kirby-Bauer disc diffusion method.

In this present study out of 108 samples studied, 84 (84%) yielded growth, while 24(22.22%) were sterile, out of 84 culture positive, 49 (58.33%) were showed monomicrobial and 35(41.66%) were showed polymicrobial growth.

The infection was most common within 20 years of age. The incidence of the CSOM decreased with increase of age.

Males were more commonly affected than females. Male: Female ratio was 2:1. Majority of the patients were belongs to lower socio economic status. Right ear was predominantly involved.

Among the aerobic bacteria majority of the isolates were gram negative bacilli. A total number of 10 different aerobic bacterial species were isolated. The most common aerobic bacteria was *Pseudomonas aeruginosa*(40%) followed by *Staphylococcus aureus* (25%).

Among anaerobic bacteria a total number of 8 different bacteria were isolated. The most common anaerobic bacteria was *Finegoldia magna* (25%) followed by *Fusobacterium necrophorum*(18.75%)

A total number of 6 different types of fungal isolates were isolated. *Aspergillus niger* (35%) was the most predominant fungus followed by *Candida albicans* (25%).

Gram positive cocci were highly sensitive to Vancomycin, Teicoplanin, Tetracycline. Gram negative bacilli belongs to Enterobacteriaceae family were highly sensitive to Ciprofloxacin and Cefepime. Gram negative non fermentative bacilli / coccobacilli were highly sensitive to Amikacin, Ticarcillin Clavulanic acid.

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