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THE PREVALENCE AND ANTIMICROBIAL RESISTANCE OF *PSEUDOMONAS* SPECIES IN PATIENTS WITH CHRONIC SUPPURATIVE OTITIS MEDIA

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Background: Chronic suppurative otitis media (CSOM) is the result of an initial episode of acute otitis media and is characterized by a persistent discharge from the middle ear through a tympanic perforation for at least 2 weeks duration. It is an important cause of preventable hearing loss, particularly in the developing world.

Methods. 1. To get an overview on the bacterial ear infection profile in general

2. To assess the antibiotic resistance of *Pseudomonal* infection (PS) particularly since it is usually the commonest infection to cause otitis media and the most difficult to treat due to the problem of multi drug resistance..

A cross sectional study was done which included 405 patient of CSOM patients. 196 (48%) case were males ,209 (52%)case were females. Swabs for aural discharge was taken from those patients. Discharge is cultured by inoculating it into blood , MacConkey agar , chocolate agars and Sabouraud agar (for fungi).

If the isolate shows to be *Pseudomonas* isolate growth then another culture of the isolate is performed on Muller Hinton Agar. Then the antibiotic susceptibility and resistance of *Pseudomonas* isolate is assessed by (Kirby-Bauer Method)

Results: 196 (48%) case were males.

209 (52%) case were females with a male to female ratio 1:1.1

150 (73%) cases were infected with *Pseudomonas* species (PS)

The sensitivity of the Pseudomonas isolates to the following antibiotics is shown below:

Amikacin 91.7%
Imipenem 89.7%
Ceftazidime 81.8%
Ciprofloxacin 73.7%
Garamycin 72.9%
Tobramycin 67.7%
Ticarcillin 66.7%
Cefoperazone 42.9%

Conclusions: *Pseudomonas* species is the commonest microorganism in cases of CSOM.

Microbiological identifications and antibiotic resistance determination of pathogens isolated from the middle ear in patients with CSOM not responding to empirical antibiotic treatment gives possibility of the choice of an effective antibiotic and its proper dosage. Cefoperazone ,a relatively new antibiotic that is used in Iraq to combat pseudomonal infections has proven to be poorly effective compared with other previously used antibiotics.

Introduction: Definition of CSOM

Chronic suppurative otitis media (CSOM) is defined as a chronic inflammation of the middle ear and mastoid cavity, which presents with recurrent ear discharges or otorrhoea through a tympanic perforation ([i]).

1. The disease is the residue of an acute suppurative infection usually acquired in infancy or early childhood ([ii]).

The disease commonly starts with painless perforation of the ear drum and purulent discharge. The perforation is usually central and often large. It remains for several years. During this time there are episodes of painless discharge of foul-smelling pus associated with a blocked ear canal and poor hearing. Between these episodes, the perforation remains, but the ear is usually dry and hearing can be normal or at least adequate ([iii]).

Generally, patients with tympanic perforations which continue to discharge
mucoid material for periods of from 6 weeks to 3 months, despite medical treatment, are recognized as CSOM cases. The WHO definition requires only 2 weeks of otorrhoea (1), but

otolaryngologists tend to adopt a longer duration, e.g. more than 3 months of active disease ([iv]).

Risk factors for CSOM

Inadequate antibiotic treatment, CSOM were previous tympanostomy tube insertion ; having had more than 3 upper respiratory tract infections and/or acute OM in the past 6 months ; having parents with a low education level and having older siblings([v]),nasal disease ([vi]), poor access to medical care ([vii]) ,bottle-feeding ([viii]), passive exposure to smoking ([ix]), attendance in congested centers such as day-care facilities ([x]), and a family history of otitis media are some of the risk factors for otitis media ([xi],[xii]). The predisposition of certain races, such as the South-western American Indians ([xiii]), Australian Aborigines([xiv]), Greenlanders, and Alaskan Eskimos([xv]) , to CSOM is also well documented. These risk factors probably favor the development of CSOM by weakening the immunological defenses, increasing the inoculum, and encouraging early infection ([xvi]).

Poor housing, hygiene and nutrition are associated with higher prevalence rates, and improvement in these aspects was found to halve the prevalence of CSOM in Maori children between 1978 and 1987 ([xvii]). Proximity to a health care facility significantly reduced the otitis media attack rate among Arizona Indian children living in reservations ([xviii]).

Bacteriology of CSOM

In CSOM the bacteria may be aerobic (e.g. *Pseudomonas aeruginosa, Escherichia coli, S. aureus, Streptococcus pyogenes, Proteus mirabilis, Klebsiella* species) or anaerobic (e.g.*Bacteroides, Peptostreptococcus, Proprionibacterium*) ([xix],[xx]).;The bacteria are infrequently found in the skin of the external canal, but may proliferate in the presence of trauma, inflammation, lacerations or high humidity ([xxi]). These bacteria may then gain entry to the middle ear through a chronic perforation ([xxii]). The most common organisms are *Pseudomonas spp.* which rapidly develop resistance to antibiotics ([xxiii]) among these bacteria, *P. aeruginosa* has been particularly blamed for the deep-seated and progressive destruction of middle ear and mastoid structures through its toxins and enzymes([xxiv])

Histopathological features of CSOM

Otitis media presents an early acute phase, with essentially reversible mucosal and bony pathological changes, which continues to a late chronic phase with well established, intractable mucoperiosteal disease. The recurrent episodes of otorrhoea and mucosal changes are characterized by osteoneogenesis, bony erosions, and osteitis that include the temporal bone and ossicles ([xxv])

The mucous membrane may be thickened by edema and submucous fibrosis and infiltration with chronic inflammatory cells. Mucosal edema may proceed to the formation of polyps. Persistent suppuration may proceed formation of granulation tissue and bone resorption. In more advanced cases, ostietis of ossicles, masoid bone and labyrinth ([xxvi]).

Complications of CSOM

Extracranial complications occur most commonly, and include mastoiditis, cholesteatoma, permanent hearing loss, paralysis of facial nerve. Intracranial complications are less common, and include meningitis, brain abscess, and hydrocephalus and lateral sinus thrombosis([xxvii]). In Australia, approximately 60% of extracranial and intracranial complications of otitis media occur in children([xxviii]).

Methods: A cross sectional study was done which included 405 patient of CSOM patients (patients with aural discharge for 2 weeks or more). The patients came to the ENT consultant clinic in Al-Khadimiya Teaching Hospital in the period 2008-2011. Swabs for aural discharge were taken from those patients from which a smear is stained with gram stain and examined by light microscope.

Also the discharge is cultured by inoculating it into blood, MacConky and chocolate agars then were incubated for 24 hr to show the responsible micro-organisms. Another inoculation into Sabouraud agar (for fungi) and incubated for 48 hr. If the growth shoes a pseudomonal growth this is verified using Gram stain and colonial morphology, motility and oxidase test.

Colonies which displayed a positive oxidase reaction were subcultured,

on Muller Hinton Agar for the purpose of assessing the antibiotic susceptibility and resistance using the Disk Diffusion Susceptibility Testing (Kirby-Bauer Method). After incubation at 35C for 16-18 hrs, zone size was measured ([xxix]) and interpreted according the CLINICAL AND LABAROTARY STANDARDS INSTITUTE (CLSI).

The antibiotic susceptibility and resistance is assessed by (Kirby-Bauer Method) by using commercially obtained disks for the following antibiotics:

The following antibiotic disks were produced by BIOANALYSE company:imipenem (IM 10ug/disk) , pipracillin

(PIP100ug/disk), cefipime (CP 30ug/disk),cefotaxime (CE 30ug/disk),ceftriaxone (CTX 30ug/disk)

carbinicillin (CAB 100ug/disk) , ticarcillin (TIC 75ug)

The following antibiotic disks were produced by Al-Raze Center for Research and Medical Therapeutic Production: tobramycin (TB 10ug) , ciprofloxacin(CIP5ug) ,amikacin(AK 30ug), garamycin(GR 10ug)

The following antibiotic disks were produced by BD company. aztreonam(ATM 30ug), ceftazidime (CZ 30ug)

Cefoperazone (CEP 75ug) containing disks were prepared in the laboratory. Appropriate concentration of cefoperazone (1 gm Cefobid [trade name for cefoperazone] vial source manufactured by Pfizer) was diluted in appropriate volume of sterile distilled water and incubated with disks which were made of filter paper .The disks were allowed to dry for 2 hr at room temperature and were stored under desiccation at -40c°.

Statistical Analysis: demographic data were demonstrated using percentages for male to female ratio.

Prevalence of the pathogenic micro-organisms which were discovered and sensitivities of the tested antibiotic disks were shown by percentages.

RESULT:

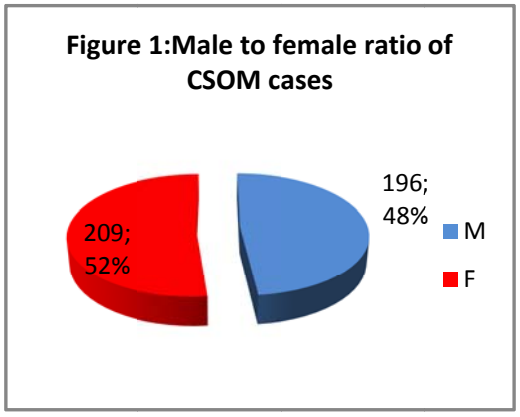
Over a period of 3 years (between march 2008 till march 2011), a total of

405 case of chronic otitis media were collected in the years 2008-2011 (Figure 1).

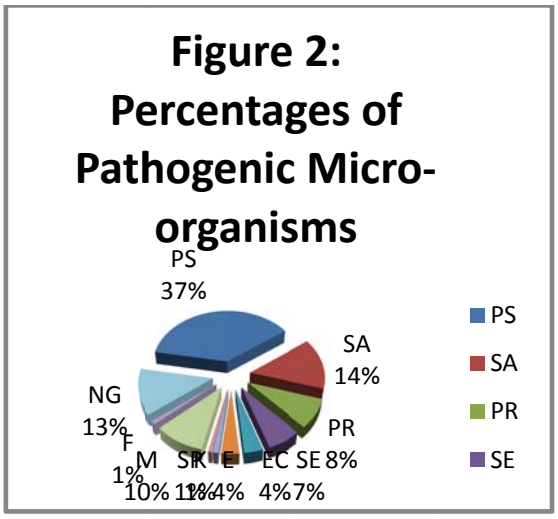
196 (48%) case were males.

209 (52%)case were females

with a male to female ratio 1:1.1

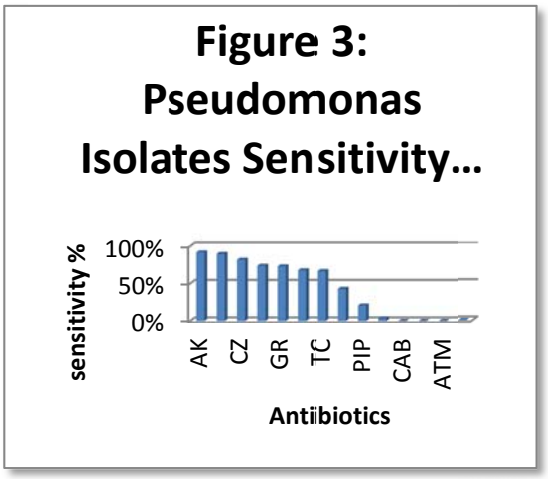


Regarding the pathogenic micro-organisms as found by the culture method (Figure 2):
150 (37%) cases were infected with *Pseudomonas* species (PS)
58 (14.3%) cases were infected with *Staphylococcus aureus* (SA)
34 (8.3%) cases were infected with *Proteus* species (PR)
27 (6.6%) cases were infected with *Staphylococcus epidermidis*(SE)
15 (3.7%) cases were infected with *Echirchia coli* (EC)
14 (3.4%) cases were infected by *Enterobacter* (E)
4 (0.9%) cases were infected by *Klebsiella* (K)
3 (0.7%) cases were infected by *Streptococcus pneumonie* (SP)
4 (0.9%) cases were infected by fungi (F)
40 (9.8%) case there was mixed infection (M)
53 (13%) case there was no growth (NG)



Regarding the sensitivity of *Pseudomonas* species isolates to the following antibiotics it is shown below (Figure 3):

Amikacin (AK) 91.7%
Imipenem (IM) 89.7%
Ceftazidime (CZ)81.8%
Ciprofloxacin (CIP) 73.7%
Garamycin (GR) 72.9%
Tobramycin (TB) 67.7%
Ticarcillin (TIC) 66.7%
Cefoperazone (CEP) 42.9%
Pipracillin (PIP) 20.7%
Ceftriaxone (CTX) 3.4%
Carbinicillin (CAB) 0.0%
Cefotaxime (CE) 0.0%
Aztreonam (ATM) 0.0%
Cefipime (CP) 0.0%



Discussion

The male to female ratio in this study is 1:1.1 .Compared to another study which has showed that the ratio is 1:1.7 ([xxx]).
Generally no gender bias is obvious in previous studies and it can occur in any age group([xxxi]).
The results show clearly that *Pseudomonas* species infections are the commonest constituting 37% of all cases .In addition to that pseudomonas species infections are the most virulent regarding the rapidity of progress and the severe complication that may happen due to the deteriorating global problem of multi drug resistance . Todar described *Pseudomonas* as being notorious for its resistance to antibiotics and for this, it is a dangerous and dreaded pathogen. ([xxxii]) The species Pseudomonas comprises of more than 140 species, only few of these are pathogenic to man like *P. aeruginosa*, *P. oryzihabitans*, and *P. plecoglossicida.*([xxxiii]) but pseudomonas aeroginosa is by far the most frequent cause and the most important pathogen in the species ([xxxiv]).
The others are essentially saprophytic and occur widely in nature ([xxxv]).

1. Other bacteria found by culture are: (14.3%) 58 cases *Staph. aureus* ,34 (8.3%) cases *Proteus* species ,27 (6.6%) cases *Staph. Epidermidis*,15 (3.7%) cases. Although *Staph. Epidermidis* is well known as normal flora of the external ear ,it can sometimes become pathogenic causing otitis media especially when the defense mechanisms are weak ([xxxvi]).

2. .*E. coli* ,14 (3.4%) cases *Enterobacter*,4 (0.9%) cases *Klebsiella*,3 (0.7%) cases *Strep. pneumonie* ,4 (0.9%) cases fungi ,40 (9.8%) case mixed infection ,53(13%) case no growth .
3. While in a study done in Basrah 2005, results were as follows: Staphylococcus aureus 25%, Pseudomonas 18.8% Streptococcus pneumonia 18.8%,Proteus sp. 8.3%,Klebsiella sp. 6.3%,E. coli 4.2%, No growth 18.8%([xxxvii]).
4. In Nigeria a study showed that : Pseudomonas 30 (44%), Staphylococcus 18 (27%), Proteus 12 (18%), Klebsiella species 8 (12%), and (0.02%) 2 were incidental fungal isolates (Candida species). 8 (12%)were contaminants while 10(15%) did not have any growth at all([xxxviii]).
5. Amikacin in this study is the most effective antibiotic in terms of sensitivity 91.7% .this finding coincides with that of a study in singapore ([xxxix]) 90.4%, and
6. Spain 91% ([xl]).
7. Cefoperazone a third generation cephalosporin has been introduced in iraq relativlely recently (in the last few years) to combat the problem of emerging drug resistance.
8. Antibiotic sensitivity regarding cefoperazone was 42.9%. In a study done in India the sensitivity was 55.87%([xli]).In another study done in Lithuania the sensitivity was 31.7%([xlii]). In a study done in Bulgaria the sensitivity for cefoperazone was 15 % ([xliii]).

Conclusions:

Pseudomonas infection in chronic otitis media is a major health problem due to its high prevalence and resistance to many drugs . Even some relatively newly developed and used drugs like cefoperazone are not very effective in combating the rapidly emerging problem of multidrug resistance.

New researches should be done to explore in depth the mechanisms of drug resistance leading to concerted efforts on the way of developing new antibiotic drug therapies.

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References:

i. WHO. Chronic suppurative otitis media .Geneva. Switzerland;2004:9
ii. Roger F.Synopsis of Operative ENT Surgery.Butterworth;1992:112
iii. Alan E Dugdale .Management of chronic suppurative otitis media.MJA 2004; 180 (2): 91-93
iv. Goycoolea MV, Hueb MM, Ruah C. Definitions and terminology. Otolaryngol Clin North America. 1991; 24 (4): 757-761.
v. Erwin L. van der Veen, Anne G. M. , et al. Predictors of Chronic Suppurative Otitis Media in Children.Arch Otolaryngol Head Neck Surg. 2006;132:1115-1118.
vi. Gibson PG, Stuart JE, Wlodarczyk J,et al.Nasal inflammation and chronic ear disease in Australian Aboriginal children. J Paediatr Child Health.1996; 32: 143-147
vii. Jahn AF. Chronic otitis media: diagnosis and treatment.Med Clin North America. 1991;75 (6): 1277-1291.
viii. [1] . Wintermeyer SM, Nahata M. Chronic suppurative otitis media. Annals Pharmacother.1994; 28: 1089-1099.
ix. Adair-Bischoff CE, Sauve RS. Environmental tobacco smoke in middle ear disease in preschool age children. Arch Pediatr Adolesc Med. 1998; 152 (2): 127-133.
x. . Fliss DM, Shoham I, Leiberman A, Dagan R. Chronic suppurative otitis media without cholesteatoma in children in Southern Israel: incidence and risk factors. Pediatr Infect Dis J.1991; 10: 895-899.
xi. Homoe P. Otitis media in Greenland. Studies on historical, epidemiological, microbiological and immunological aspects. Int J Circumpolar Health. 2001; 60 (Suppl 2): 1-54.
xii. Kenna MA. Treatment of chronic suppurative otitis media. Otolaryngol Clin North Am.1994; 27 (3): 457-472.
xiii. Wiet RJ. Patterns of ear disease in the Southwestern American Indian. Arch
xiv. Otolaryngol. 1979; 105: 381-385
xv. Sunderman J, Dyer H. Chronic ear disease in Australian aborigines.Med J Aust. 1984;140: 708-711
xvi. Maynard JE, Fleshman JK, Tschopp CF. Otitis media in Alaskan Eskimo children.JAMA.1972; 219: 597-599.
xvii. Berman S. Otitis media in children. N Eng J Med. 1995; 332 (23): 1560-1565
xviii. World Health Organization. Prevention of hearing impairment from chronic otitis media. Report of a WHO/CIBA Foundation Workshop. Geneva. 1998.
xix. Wiet R, DeBlanc G et al. Natural history of otitis media in the American native. Ann Otol Rhinol Laryngol. 1980; 89: 14-19.
xx. Brobby GW. The discharging ear in the tropics: a guide to diagnosis and management in the district hospital. Tropical Doctor. 1992; 22 (1): 10-13.
xxi. Brook I, Frazier E. Microbial dynamics of persistent purulent otitis media in children. J Pediatrics. 1996; 128(2): 237-240.
xxii. [1] . Mawson S, Pollack M. Special role of *Pseudomonas aeruginosa* in chronic suppurative otitis media. Ann Otol Rhinol Laryngol Head and Neck Surg. 1988;97 (Suppl. 130):10-13.
xxiii. otitis media. Arch Otolaryngol Head Neck Surg. 1988; 97 (2) (Suppl. 137): 16-17.
xxiv. Alan E Dugdale .Management of chronic suppurative otitis media.MJA 2004; 180 (2): 91-93
xxv. WHO. Chronic suppurative otitis media .Geneva. Switzerland;2004:10
xxvi. Meyerhoff W. Pathology of chronic suppurative otitis media. Ann Otol Rhinol Laryngol Head and Neck Surg. 1988; 97 (Suppl. 130): 21-24.
xxvii. Cuneyt M. Alper, Charles D. Advanced therapy of otitis media. PMPH-USA.2004; page 262

xxviii. Kangsanarak J, Fooanant S, Ruckphaopunt K, et al. Extracranial and intracranial complications of suppurative otitis media. Report of 102 cases.

xxix. O'Connor TE, Perry CF, Lannigan FJ. Complications of otitis media in Indigenous and non-Indigenous children.Med J Aust. 2009;191(9 Suppl):S60-4.

xxx. ChingChingWu.Disk Diffusion Susceptibility Testing (Kirby-Bauer Method)[internet] . ADDL .1997 .[cited 2011 April 5]. available from: http://www.addl.purdue.edu/newsletters/1997/spring/dds.shtml

xxxi. AS Adoga et al. Swab and aspiration specimen collection methods and antibiogram in chronic suppurative otitis media at Jos University Teaching Hospital: Which is superior?. Department of Surgery, Ear, Nose and Throat unit, Jos University Teaching Hospital. Ibadan. Nigeria. 2010; 9(4): 230-234

xxxii. Cholesteatoma (chronic suppurative otitis media): Treatment, symptoms, advice and help[internet][cited on 22/4/2011]. available from : http://www.privatehealth.co.uk/diseases/ear-nose-throat/cholesteatoma/

xxxiii. Kenneth Todar .Pseudomonas aeruginosa[internet].Todars online textbook of bacteriology [cited on 23/4/2011].available from;http://www.textbookofbacteriology.net/pseudomonas.html

xxxiv. Pseudomonas_aeruginosa[internet].Wikepedia The Free Encyclopedia.[cited on 23/4/2011]. available from:http://en.wikipedia.org/wiki/Pseudomonas_aeruginosa

xxxv. [1] . Luis M. ,De la Maza . Pseudomonas .Color atlas of medical bacteriology. ASM Press; 2004:133

xxxvi. Cruickshank, R.J.P., Duguid, B.P. Medical Microbiology. Churchill Living Stone Edinburgh London and New York. 1975; : 195-200.

xxxvii. harles D, Bluestone.staphylococcus epidermidis otitis media pathogenic. Pediatric otolaryngology. Elsevier Health Science.٢٠٠3;551

xxxviii. *Ahmed M. Al-Abbasi* .Efficacy of Povidone Iodine in Treatment of Active Chronic Suppurative Otitis Media. JIMA.2006;(38) 118-121

xxxix. S Adoga et al. Swab and aspiration specimen collection methods and antibiogram in chronic suppurative otitis media at Jos University Teaching Hospital: Which is superior?. Department of Surgery, Ear, Nose and Throat unit, Jos University Teaching Hospital. Ibadan. Nigeria. 2010; 9(4): 230-234

xl. hean Yen Tan, Li Yang Hsu, Tse Hsien Koh,et al. Antibiotic Resistance in Gram-negative Bacilli: A Singapore Perspective. Acad Med Singapore 2008;37:819-25

. E. Bouza, F. Garcia-Garrote, E. Cercenado, et al . Pseudomonas aeruginosa: a Survey of Resistance in 136 Hospitals in Spain. Antimicrobial Agents and Chemotherapy.1999;43(4) 981-982.

xli. S Shenoy, S Baliga. Antibiotic sensitivity patterns of Pseudomonas aeruginosa strains isolated from various clinical specimens. Medicina (Kaunas) 2007; 43(1)

xlii. he peculiarities of *Pseudomonas aeruginosa* resistance to antibiotics and prevalence of serogroups
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Department of Microbiology, Kaunas University of Medicine, Lithuania

xliii. Tanya Strateva, Vesselа Ouzounova-Raykova, Boyka Markova,et al. Problematic clinical isolates of Pseudomonas aeruginosa from the university hospitals in Sofia, Bulgaria: current status of antimicrobial resistance and prevailing resistance mechanisms.J Med Microbiol .2007;56 : 956-963