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OTITIS EXTERNA: BACTERIAL ETIOLOGY, AGE-WISE AND GENDER-WISE RELATEDNESS

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Abstract: **:** In India, most of the study in case of ear infection is done on otitis media but otitis externa is also one of the main causes of ear infection which may lead to impairment of hearing. Otitis externa is the infection and inflammation of the external ear and ear canal up to eardrum. It is also called swimmer's ear as it is always associated with the swimmers. Present study concerns with the isolation and identification of bacterial pathogens from the patients of otitis externa and age-wise and gender-wise distribution of patients. The samples were collected by using a standard collection method from the infected ear and processed in the laboratory for the isolation of pathogenic bacteria. The bacteria were identified on the basis of 16SrRNA gene sequencing and phylogenetic analysis. A total of six bacterial species was identified as Staphylococcus, Klebsiella, Bacillus, Acinetobacter, Exiguobacterium, and Enterococcus. Staphylococcus was found as the most frequently isolated bacterial species. In an age-wise distribution study out of 80, 43 patients belonged to the age group 0-18 years, 35 from the age group above 40 years, and 2 from the age group 19-40 years. In the gender-wise distribution study from a total of 80 patients, 57 were females and 23 were males. The most affected age group of otitis externa is children while the females were more affected than males. The high incidence of Staphylococcus showed the commonest cause of otitis externa. The data regarding isolated pathogens will be useful for topical antibiotic treatment.

Keywords: Otitis externa, causative agents, *Staphylococcus*, Swimmer's ear

I. INTRODUCTION

Like skin external ear is always exposed to various micro flora and factors like moisture, amount and composition of cerumen, pH and trauma to the skin decides the health of external ear. Otitis externa is a generic term for inflammation of the external auditory meatus (EAM) skin, which includes not only the ear pinna but also the portion of the ear canal that leads up to the eardrum without extending to the middle ear(Enoz & Sevinc, 2009) . Otitis externa is also called as swimmer's ear. The infection is similar to infection of skin and soft tissues elsewhere. Swimming in the polluted water is the cause of swimmer's ear but water trapped in ear canal after shower also causes swimmer's ear(Mustafa et al., 2015) . Mainly there are some factors which are responsible to cause otitis externa and these are excessive moisture which increases the pH and decreases the cerumen, high humidity, warm swimming pools, trauma and use of hearing aids(Cirpaciu et al., 2017) . As the severity progresses the otitis externa classified into three types, a) acute otitis externa, b)chronic otitis externa and c)malignant otitis externa. Acute otitis externa is also known as swimmer's ear and it is defined as diffuse inflammation of the external auditory canal. It is common disease in children, adolescents and adults associated with swimming(Hui et al., 2013) . Chronic otitis externa is characterized as thickening of the EAC (external auditory canal) skin secondary to low grade infection and inflammation. It is caused by irritation from drainage through a perforated tympanic membrane. Malignant external otitis infection begins as an inflammation of the external auditory canal that later spreads to the deeper structures, such as cartilage and bone of the skull base. It affects diabetics and immunocompromised people(Bhat et al., 2015) .

The aim of the study was to isolate and identify the bacterial species responsible for the infection of external ear. The study also focused on deviation in the number of patients from the different age groups and also gender of patients.

Figure1Patients showing infection of external ear

II.MATERIAL AND METHODS

Present study was conducted from December 2018 to October 2019 with total 80 cases of clinically diagnosed otitis externa selected from the Urban and rural area of different places of Akola, Maharashtra (India). These total 80 samples were divided according to the types of otitis externa i.e. acute otitis externa, chronic otitis externa and malignant otitis externa.

The scrapping and swabbing samples were collected using standard microbiological techniques in the sterilized collection vials containing normal saline solution as the transport medium and transported to the laboratory within 1 hour (as soon as possible) of collection and processed for further isolation and identification of pathogenic bacteria. The propagation was done by inoculating the collected samples into the sterilized nutrient broth and inoculated for one day at 37^0C. MacConkey agar (for Gram negative bacteria)(Ghanpur et al., 2017) and Mannitol salt agar (for Gram positive bacteria) of HiMedia laboratory were used as a growth media for culturing of samples and incubated at 37^0C for 24hrs to get the proper growth of bacteria. Identification of isolated pathogens was done by using Gram staining technique, standard biochemical methods(Reyes, 2018) (Table.1), 16SrRNA gene sequencing and phylogenetic analysis(Castro-Escarpulli et al., 2015) (Fig.3). The 16SrRNA gene sequencing and phylogenetic analysis was done at geneombio TECHNOLOGIES PVT. LTD.VEDANT, S. No. 39/3, H. No. 1043, Yogi Park, Off Mumbai Bangalore Expressway, Baner, Pune 411045. Maharashtra, INDIA. All the routine was carried out by following the standard microbiological techniques and work was carried out with the approval from Institute ethical committee.

III. RESULTS

In the present study a total 80 samples were processed in the laboratory for the isolation and identification of pathogenic bacteria. In case of age-wise distribution study from total 80 patients 43 patients were from the age group 0-18years, 35 patients from the age group above 40years and very less patients i.e. only 2 from age group 19-40years (Table1). In the gender-wise distribution study from total 80 patients 57 were females and 23 were males.

Total 124 bacteria were isolated from the collected samples and from these 124 isolates six bacterial strains (nearest neighbor) were identified as (PPM1A) *Klebsiella pneumoniae* strain CIFRI-OBBKP2, (PPM2B) *Staphylococcus simiae* CCM 7213, (PPM2C) *Bacillus subtilis* strain NCDO 1769, (PPM8A) *Acinetobacter pittii* DSM 21653 strain ATCC 19004, (PPM9A) *Exiguobacterium acetylicum* strain DSM 20416 and (PPM9B) *Enterococcus mundtii* strain NBRC 100490 on the basis of phylogenetic analysis (Figure2). Among 124 isolated species 39 (31%) were of *Staphylococcus* sp., 28 (23%) were of *Klebsiella* sp., 21 (17%) were of *Bacillus* sp., 15 (12%) were of *Exiguobacterium* sp., 12(10%) were of *Acinetobacter* sp. and only 09 (7%) were of *Enterococcus* sp. (Figure 3).

Table 1Age-wise distribution of number of samples and number of isolates of respective samples

Age group	Number of samples	Number of isolates
0-18yrs	43	69
19-40yrs	02	04
Above 40yrs	35	51
Total	80	124

Table 2Morphological and biochemical characteristics of isolated pathogens

Bacterial isolates (code)		PPM1A	PPM2B	PPM2C	PPM8A	PPM9A	PPM9B
Gram Characteristics		-	+	+	-	+	+
Morphological characteristics	Shape	Rods	Cocci	Rods	Coccobacilli	Rods	Cocci
	Motility	NM	NM	M	NM	M	NM
Biochemical characteristics	Indole	-	-	-	-	-	-
	MR	-	+	-	-	-	-
	VP	+	+	+	-	+	+
	Citrate	+	-	+	+	+	+
	Glucose	+	+	+	+	+	+
	Lactose	+	+	+	+	-	+
	Mannitol	+	+	+	-	+	+
	Catalase	+	+	+	+	+	+
	Oxidase	-	-	V	-	-	+
	DNase	-	+	-	+	+	+
	Coagulase	-	+	-	-	-	-
	Urease	+	+	-	V	-	V
	Gelatinase	-	-	+	+	+	-

-: Negative, +: Positive, M:Motile, NM: Non motile, V: Variable, MR: Methyl red, VP: Voges Proskauer, PPM1A, PPM2B, PPM2C, PPM8A, PPM9A and PPM9B: codes of isolated bacteria

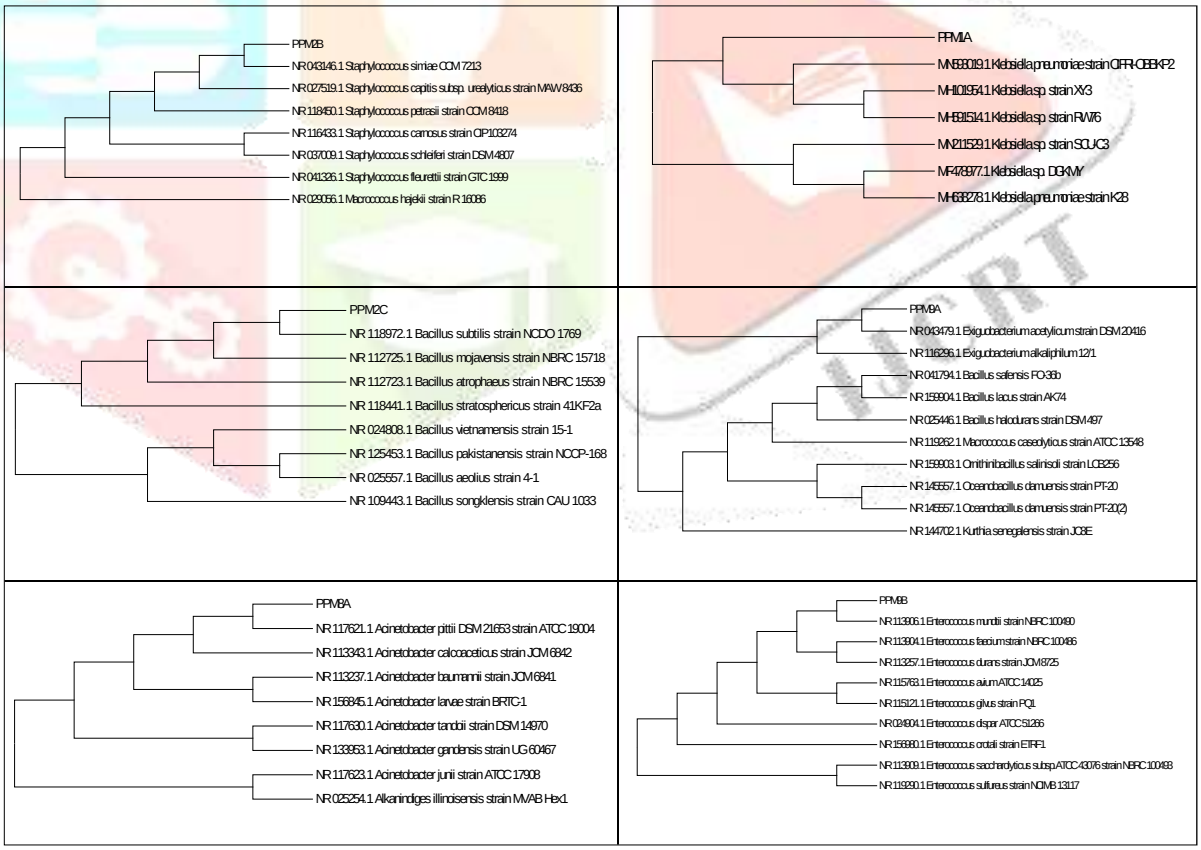

Figure 2 Phylogenetic trees of isolated pathogenic bacteria from external otitis

Codes of isolated bacteria and its nearest neighbor in bracket: PPM1(*Klebsiella pneumoniae* strain CIFRI-OBBKP2), PPM2B(*Staphylococcus simiae* CCM 7213), PPM2C(*Bacillus subtilis* strain NCDO 1769), PPM8A(*Acinetobacter pittii* DSM 21653 strain ATCC 19004), PPM9A(*Exiguobacterium acetylicum* strain DSM 20416) and PPM9B(*Enterococcus mundtii* strain NBRC 100490).

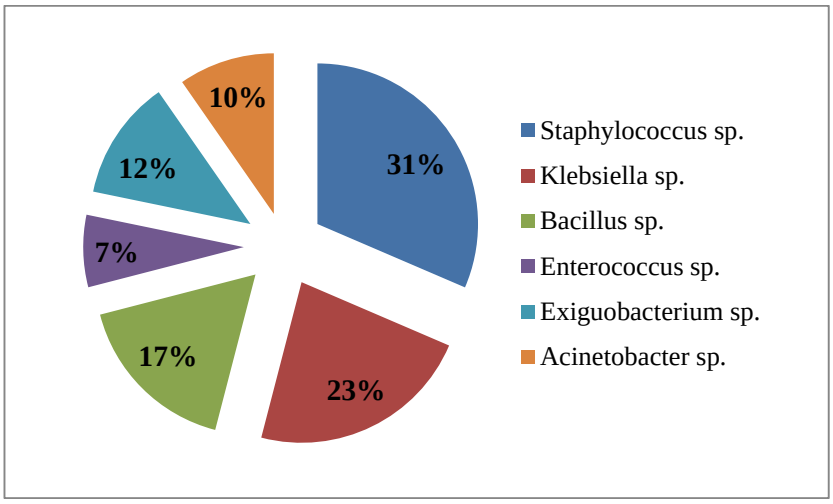

Figure 3 percent distribution of bacterial isolates from samples of external otitis

IV. DISCUSSION

In the present study considering all age group the female were more affected than male which is contrary to the study in which from total 64 patients 34 were male and 30 were female(Ghanpur et al., 2017) , same results were also found in another study(Cirpaciu et al., 2017) where from 59 patients 35 were male and 24 were female. Again one study revealed that out of 42 samples which was positive for *Pseudomonas aeruginosa*, gender-wise distributions were higher for female as compared to males, 22 samples (52%) were positive for females and for male 20 samples (48%) were found positive(Khattak et al., 2013) . Present study reveled that as 43 samples out of 80 samples were from the age group 0-18 yrs i.e. more than 50% samples, which reflected the most affected age group.

Among all the isolated pathogenic bacteria 31% was Staphylococcus strain which showed significantly high incidence followed by Klebsiella, Bacillus, Exiguobacterium, Acinetobacter and Enterococcus respectively. got similar results that 57% of all cases the high incidence was of *Staphylococcus aureus*(Cirpaciu et al., 2017) . was found similar results, high percentage of *Staphylococcus aureus* (37%), *Pseudomonas aeruginosa* (29.6%) followed by other organisms(Nogueira et al., 2008) . The most common bacteria in the patients of otitis externa were *Staphylococcus aureus* (20.8%), *Bacillus* (18.9%) and *Pseudomonas* (11.3%) studied by(Kiakojori et al., 2010).

V. CONCLUSION

The data in the present study concluded that the most affected age group was children, which may be due to children are more vulnerable than adults to environmental risks. Females were most affected considering the gender. The high incidence of *Staphylococcus* gave alarming sign and the data regarding the common pathogens is useful while treating the otitis externa. Further emphasis on antibiotic study will provide a way of promising topical antibiotic treatment to otitis externa.

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