**The isolation of Proteusspecies from patients**

**with otitis media in Baquba city**

**Abass A. Al-Duliami (Ph.D.), college of Education, Diyala University**

**Abdul-Razak SH. Hasan (Ph.D.), College of Medicine, Diyala University**

**Kariem S. Al-Ajeeli (Ph.D.) College of Vet. Medicine, Diyala University**

**Afak Rashid (M.Sc.), College of Education, Diyala University**

**Abstract**

**Background:**

Otitis media is an infection of the middle ear, usually associated with upper respiratory tract infection.Although, it can occur at any age, but 60-80% of children had recurrentotitis media**.** Early diagnosis and management may avoid serious complications like, brain abscess, meningitis, lateral sinus thrombosis and subdural abscess**.**

**Objectives:**

To determine the isolation rate of proteus species from patients with otitis media in Baquba city, and to explore the effects of certain relevant factors.

**Materials and methods:**

Ear swabs were collected from 270 patients with acute or chronic otitis media attending the out-patient clinic of Baquba General Hospital.Information regarding age, sex, residence, and smoking were collected by special form pre-constructed for this purpose. Isolation and identification of Proteus species based on standard bacteriological criteria. All data were statistically analyzed.

**Results:**

Proteus species were isolated from 35(12.9%). The *P.mirabilis* constitute 28 and *P.vulgaris* 7. Bacterial growth other than proteus constitute 75.9%, while swabs with no bacterial growth was 11.1%. The mean age of patients was 25.9±16.8. The isolation rate from males (17.8%) was higher than that of females (8.5%). The highest isolation rate (18.4%) was found in 10-19 years age group, while the lowest isolation rate (6.6%) was recorded in those <5 years old. Residents of rural areas recorded highest number of *Proteus species* isolates when compared to those of urban areas (13.3% vs 11.9%). Chronic OM cases showed higher isolation rate (18.6%) of *Proteus spp* than that of acute cases (6.9%). Additionally, the isolation rate (21.1%) of *Proteus spp*. from smokers with OM was higher than that of negatively smokers (14.6%) and non- smokers (9.9%).

**Conclusion:**

Higher isolation rate of Proteus species were recovered from patients with chronic otitis media.

**Keywords: Otitis media, Proteus, Upper respiratory tract nfection**

**Introduction**

Otitis media, an infection of the middle ear, is usually associated with upper respiratory tract infection. Although, it can occur at any age, but 60-80% of children infected with recurrent otitis media **[1]**. Early diagnosis may avoid the possibility of complications associated with such infection like, brain abscess, meningitis, lateral sinus thrombosis and subdural abscess **[2]**. The infection was attributed to many bacterial species like *Staphylococcus aureus, Proteus mirabilis, Proteus vulgaris, and Pseudomonas aeruginosa* **[3,4].** Van Hasselt & Van Kregten (2002) **[5]** found that most cases in children in Africa associated with fecal bacteria, and that 74% of the isolates were *P. mirabilis* and 60% were *Enterococci spp*. Many researchers isolated some other bacteria such as *Staphylococcus epidermidis* **[6]**, *Streptococcus pneumoniae* **[7]**, *Pseudomonas aeruginosa* **[8,9]**, and *Moraxella catarrhalis* **[10]**. Cases associated with fungus infection such as *Aspergillus spp*. and *Candida albicans* were also reported **[2]**. This study was conducted, firstly to screen proteus species as one of these causative agents of otitis media patients in Baquba and its peripheries, and secondly to study the effect of some host factors like gender, age, residence, severity of the disease and smoking habit on the isolation rate of proteus species isolates from patients with otitis media.

**Patients and Methods**

This present study was conducted on 270 patients with otitis media attended outpatient clinic of Baquba General Hospital. The patient group includes 141 females and 129 males, 130 of them with acute otitis media and the remaining 140 with chronic otitis media. The age range was less than 5 to more than 50 years.

Sterile bacteriological swabs were used to collect middle ear effusion from patients. Swabs were directly streaked on blood agar, MacConkey agar, and chocolate agar plates. Plates were incubated at 37 °C for 18-24 hrs. Bacterial colonies on the culture media were selected and identified according to standard bacteriological and biochemical criteria (Collee et al., 1996) **[11]**. For proteus speces colonies with swarming phenomenon were selected, stained with gram stain and microscopically examined. Furthermore, fresh same colonies were inoculated on blood agar, incubated at 37 °C and identified as above.

**Results and Discussion**

The patient group includes 141(52.2%) females with mean age of 23.8±19.9 years, and 129 (47.8%) males with mean age of 27±6.5 years. According to the severity of the disease, 130 (48.1%) of patients were with acute OM, and 140 (51.9%) patients with chronic disease. This study revealed that 203 (75.2%) patients were from rural areas and 67 (24.8%) were from urban areas. Looking through their smoking habit, 57 (21.1%) individuals were smokers, 172 (63.7%) were non-smokers and 41(15.2%) were negatively smokers.

According to the bacteriological and biochemical criteria, the results revealed that 35 (13%) isolates were belong to proteus species; 7 (2.6%) were *P.vulgaris* and 28(10.4) were *P.mirabilis*. Additionally, 205 isolates were belonging to bacteria other than proteus species, and 30 swabs yield no bacterial growth, table (1).

**Table (1): The outcome of ear swabs cultivation.**

|  |  |  |  |
| --- | --- | --- | --- |
| Result of cultivation | | No. of isolates | (%) |
| Negative for bacterial growth | | 30 | (11.1) |
| Bacterial growth other than proteus | | 205 | (57.9) |
| Positive for proteus growth | *P.vulgaris* | 7 | (02.6) |
| *P.mirabilis* | 28 | (10.4) |
| Total Number of Isolates | | 270 | (100) |

The isolation rate of proteus species obtained in the present study was higher than that reported by other worker (Al-Saadi,2001) **[12]** who found that the isolation rate of proteus species recovered from patients with otitis media in Baghdad was 14.3%, 48 of them (13.7%) were *P.mirabilis* and 2 were *P.vulgaris*. In another study on patients with otitis media, the proteus species constitute 14.7%, while bacteria other than proteus species constitute 85.3% including *S.aureus* and *Ps. aeruginosa* **[13]** . Furthermore, 11.1% of the swabs were negative for bacterial growth. These may either due to fungal or viral infections **[3,9,14]**.

Regarding the patient gender, the results showed that the isolation rate of proteus species from male was significantly higher than thatn of female (17.8% *vs* 8.5%), table (3).

**Table (2): Isolation rate of proteus species by patient gender.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Gender | proteus  (+) | | proteus  (-) | | Total number | | P  (X2 ) |
| No. | (%) | No. | (%) | No. | (%) | 0.002 |
| Female | 12 | (8.5) | 129 | (91.5) | 141 | 100 |
| Male | 23 | (17.8) | 106 | (82.2) | 129 | 100 |

These results were in agreement with previous studies **[15]**. However, the present results were inconsistent with other workers **[10,16]**. These controversial results may be related to hormonal factors related to the sex **[17]**.

Although, the highest isolation rate of proteus was found among 10-19 years old compared to other age groups; however, the difference was statistically insignificant, table (3).

**Table (3): Isolation rate of proteus species by patient age.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Age groups (Ys) | proteus (+) | | proteus (-) | | Total number | | P  (X2) |
| No. | (%) | No. | (%) | No. | (%) | 0.44 |
| <5 | 4 | (6.6) | 57 | (93.4) | 61 | 100 |
| 5-9 | 4 | (12.6) | 28 | (87.5) | 32 | 100 |
| 10-19 | 7 | (18.4) | 31 | (81.6) | 38 | 100 |
| 20-49 | 15 | (15.3) | 83 | (84.7) | 98 | 100 |
| 50+ | 5 | (12.2) | 36 | (87.8) | 41 | 100 |

The previous studies regarding the isolation rate of proteus species according to patient age have yielded controversial results, the probable contributing factors may include geographical area, sample size and laboratory techniques employed **[16,18]**. It has been suggested that the presence of Staphylococcusspecies and/or Streptococcus spcies may limits the growth of other bacteria including the proteus species **[19]**.

The present results also revealed that there was insignificant difference regarding the isolation rate of proteus species from patients reside in rural areas compated to those reside in urban areas (11.9% *vs* 13.3%), table (4).

**Table (4): Isolation rate of proteus species by residence.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Residence | Proteus (+) | | Proteus (-) | | Total Number | | P  (X2) |
| No. | (%) | No. | (%) | No. | (%) | 0.77 |
| Rural | 27 | (13.3) | 176 | (86.7) | 203 | 100 |
| Urban | 8 | (11.9) | 59 | (88.1) | 67 | 100 |

A consistent, but higher rate of proteus species has been documented by (Hafidh *et al*., 2006) **[4]** who reported 73.3% isolation rate of *P.mirabilis* from patients belong to rural areas of Baquba. This might be due to the bad sanitary conditions in rural areas when compared to urban **[5]**. Most cases from rural areas are associated with Proteus species and *Ps. aeruginosa* isolates, where the fecal route is the main source of infection **[20]**.

The isolation rate of proteus species from cases of chronic otitis media was significantly higher than that from acute cases (18.6% *vs* 8.9%), table (5).

**Table (5): Isolation rate of proteus species by disease category.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Disease category | Proteus spp. (+) | | Proteus spp.(-) | | Total number | | P  (X2) |
| No. | (%) | No. | (%) | No. | (%) | 0.004 |
| Acute | 9 | (6.9) | 121 | (93.1) | 130 | 100 |
| Chronic | 26 | (18.6) | 114 | (81.4) | 140 | 100 |

Many other researchers recorded similar results **[1,4,21]**. Such results may be attributed to the opportunistic and ubiquitous natures of the proteus species **[2,5,22]**.

Table (6) showed that the isolation rate of proteus species from smoker patients (21.1%) was higher than that of the negatively smokers (14.6%) and non-smokers (9.9%).

**Table (6): The effect of smoking on the isolation rate of proteus species.**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Smoking Habit | Proteus (+) | | Proteus (-) | | Total Number | | P  (X2) |
| No. | (%) | No. | (%) | No. | (%) | 0.09 |
| Non-Smokers | 17 | (09.9) | 155 | (90.1) | 172 | 100 |
| Smokers | 12 | (21.1) | 45 | (78.9) | 57 | 100 |
| Negatively Smokers | 6 | (14.6) | 35 | (85.4) | 41 | 100 |

These results were concordant with previous studies **[23]**. It has been found that 61% of otitis media infections among children below two years of age exposed to household wood and charcoal smoke **[24]**. Furthermore, it has been documented that changes in epithelial cells of the respiratory tract occurred due to smoking, reduced the immune response and enhance the possibility of otitis media and meningococcal diseases **[25]**.

In conclusion, this study showed that proteus species was one of the acute and chronic otitis media causative agents in Diyala province. Furthermore, the gender, age, residence, severity of the disease and smoking habit of individuals appeared of contrary effects on the infection with these bacteria.

**References**

1.Kalcioglu,M.T. ; Ozturan,O. ; Durmaz, R. and Aktas,E. In vitro efficacy of the successive or staggered use of eardrops. Eur.Arch.Otorhinol. 2006; 263(5):395-398.

2.Pendido Nde, O. ; Borni, A. ; Iha,L.C. ; Suguri,V.M. ; Onishi,E. Itracranial complications of otitis media : 15 years experience in 33 patients. Otolaryngol.Head Neck Surg. 2005: 132(1):37-42.

3.Pajor,A. ; Durko,M. ; Jankowski,A. ; Bartoszko-Tyczkowska,A. and Stanczyk,R. Bacteriological evaluation in chronic otitis media. Otolaryngol.Pol. 2006: 60(5):757-763.

4. Hafidh,M.A. ; Keogh,I. ; Walsh,R.M. ; walsh,M. and Rawluk,D. Otogenic intracranial complications: a 7 years retrospective. Am.J.Otolaryngol. 2006: 27(6):390-395.

5. Van Hasselt,P. and Van Kregten,E. Treatment of chronic suppurative otitis media with ofloxacin in hydroxypropyl methylcellulose ear drops: a Clinical bacteriological study in a rural area of Malawi. Int.J.Pediatr.Otorhinolaryngol. 2002: 63(1):49-56.

6. Al-Saquer,R. K. A study on certain aerobic bacteria resistant to antibiotics isolated from patients with otitis media. M.Sc. thesis, College of science, Baghdad University, 2000.

7.Ako-Nai,A.K. ; Olugo,F.A. ; Onipede,A.O. ; Adejuyigbe,E.A. and Amusa,Y.B. The characterization of bacterial isolates from acute otitis media in lle-lfe, southwestern Nigeria. J.T.Pediatr. 2002: 48(1):15-23.

8. Al-Duliami, A.; Al-Jaaferi, A.H.; Nasirlleha, B.A. and l-Duliami, H.H. A bacteriological study of patients with otitis media in Baquba. Al-Fatih J.2002: 10:

9.Kuczkowski,J. ; Piatek,R. and Kur,J. Bacterial infections in chronic otitis media usefulness of molecular diagnosis based on PCR method. Otolaryngol.Pol. 2004; 58(3):497-504.

10. Al-Tarfi, B.A. Isolation and identification of some bacteria and fngi causing otitis media. M.Sc. thesis. College of Science-Al-Mustansyria University, 2002.

11.Collee,J.G. ; Fraser,A.G. ; Marmian,B.P. and Simmons,A. “Mackie and McCartney Practical Medical Microbiology”. 14th ed. 1996. Chuirchill Livingstone Inc. USA.

12. Al-Saadi, H.A. Effect of crude protein extracted from Proteus mirabilis isolated from otitis media on immunological cells. M.Sc. thesis. College of Science, Al-Mustansyria University, 2001.

13 Kovacic,M. and Dzelalija,B. Clinical success of treatment of chronic otitis media using topical and per oral administration of Ofloxacin. Lijec-Vjesn, 1999; 121(6):185-187.

14.Tong,H.H. ; Wieser,J.N. ; James,M.A. and deMaria,T.F. Effect of influenza A virus infection on nasopharyngeal colonization and otitis media induced by transparent or opaque phenotype variants of *Streptococcus pneumonia* in the cinchhilla model. Infect. Immunol. 2001; 69:602-606.

15.Pettigrew,M.M. ; Gent,J.F. ; Zhu,Y. ; Triche, E.W. ; Belanger, K.D.; *et al*. Association of surfactant protein A polymorphisms with otitis media in infants at risk for asthma. BMC Medical Genetics, 2006; 7(68):1471-2350.

16. Al-Showekh, R.M. Isolation and identification of certain bacteria causing otitis media with molecular study of some species. M.Sc. thesis. Al-Mustansyria University, 2002.

17.Morris,P.S. ; Leach,A.J. ; Silberberg,P. ; Mellon,G. ; Wilson,C. ; *et al.* Otitis media in young aboriginal children from remote communities in northern and central Australia: a cross-sectional survey. BMC Pediatr. 2005; 5:1471-2431.

18. Al-Saadi, Z.N.; Kasim, F.H. and Al-Fatah, A. Isolation and identification of bacteria causing respiratory tract infections in Tikrit city and determination of its resistance to antibiotics. Tikrit J. Pure Sci. 2005;10(1): 16-20.

19. Brooks,G.F. ; Butel,J.S. ; Caroll,K.C. and Morse,S.A. Jawetz, Melnick and Adelberg’s, Medical Microbiology. 24th ed. 2007. Appleton and Lange.

20. Van Rooty,C.H. ; Swart,J.G. ; Opthof,J. ; Vlantis,A.C. ; Ahmed,M.R. and Venter,P.H. Diagnosis and treatment of ear disease among children in the Ellisras district. An outreach programme. S.Afr.Med.J. 1995; 85(7):675-677.

21. Klein,J.O. In vitro and In vivo antimicrobial activity of topical ofloxacin and other ototopical agents. Pediatr. Infect. Dis.J. 2001; 20(1):102-103.

22.O’Hara,C.M. ; Brenner,F.W. and Miller,J.M. Classification, identification and clinical significance of *Proteus, Providencia* and *Morganella*. Clin.Microbiol.Rev. 2000; 13(4):534-546.

23. Miller,K.E. Effect of smoke exposure on otitis media recurrence. J.Am. Family Physician, 2002; 66(4).

24. Da Costa,J.L. ; Navarro,A. ; Neves,J.B. and Martin,M. Household wood and charcoal smoke increase risk of otitis media in childhood in Maputo, Inter.J.Epidemiol. 2004; 33:573-578.

25. Arcavi,L. and Benowitz,N.L. Cigarette smoking and infection. Arch.Intern.Med. 2004; 164:2206-2216.