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AN AEROBIC MICROBIOLOGICAL CORRELATION BETWEEN DISCHARGING EAR AND THROAT BACTERIAL FLORA

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Abstract :
Chronic suppurative otitis media is very common in developing countries and forms a major case load in otorhinolaryngology OPDs of hospital. We have done a study on bacteriological co-relation between discharge of ear and throat bacterial flora in normal throat or infected throat.
Study was done on 157 cases which had chronic suppurative otitis media and Gram's staining as well as culture sensitivity was done to find out the organism.
Throat swabs were taken where 52 cases had complaint of throat infection and
102 cases had normal throat. Gram positive cocci was most common bacteria on Gram's staining as well as on culture from ear swab and throat swab. Except in cases of Squamosal type of chronic suppurative otitis media where Gram negative bacilli like Pseudomonas and Klebsiella were common organism grown.
On establishing relationship statistically, study showed that there is significant relationship present in mucosal chronic suppurative otitis media and infected throat. While no relationship existed between mucosal chronic suppurative otitis media and normal throat bacterial flora.

Introduction

Chronic suppurative otitis media is a very ancient disease known to exists since Hippocrates's days who noted that "acute pain of the ear with continued high fever is to be a dreaded, for the patient may become delirious and die"[1]. It is also a very common condition encountered by clinicians in day-to-day practice specially in developing world presumably due to the poor hygienic condition, dense population and people living in poorly ventilated and congested environment. L Monasta in 2012[2], studied the global impact of CSOM and stated that incidence of acute otitis media worldwide is 10.85% with 51% occurring in less than five years old and that of chronic suppurative otitis media is 4.76% with 22.6% occurring annually in under-fives. It is estimated that twenty thousand people die each year from otitis media. Mostly prevalence of URTI predisposes the people for infection in ear. Running nose as commonly seen in lower-economic population, is commonly associated with adenoid hypertrophy in children and discharging ear. Thus, the prevalence of infection of Middle ear cleft could be due to reflux, aspiration or insufflation of bacteria into middle ear from nasopharynx[3,4]. Exudates from ear contains numerous organisms like Staphylococcus aureus, Pseudomonas aeruginosa, B. fragilis, B. catarrhalis and proteus species[5,6,7,8]. Alem Getaneh et al (2021)[9] also reported in his retrospective study a positive culture rate of 76% and presence of

Staphylococcus aureus (27.9%), *Proteus* species (20.8%), *Streptococcus* species (10%), and *Pseudomonas* sps (8.92%) as main bacterias cultured from ear swab. Paul noone[10] and Papastavros et al[11] also suggested that Staph. Epidermidis, Pseudomonas aeruginosa, & Kliebsiella species also form commensal flora of External auditory canal and acts as an opportunistic pathogens. Tonsillitis, was known even before the days of Hippocrates and when all the infection of throat and necks were known as CYNANCHE. However, Hippocrates reserved this name for the inflammation of throat only[12]. Throat infection is one of the commonest condition, which can precede or be sequalae of Upper respiratory tract infection[8]. The normal flora of throat mainly comprises of H. influenzae, Pneumococcus, Streptococci, Staphylococcus and Neisseria species. These are also seen in discharging ears especially in acute exacerbation[13].
Bacteriological co-relation between adenoids and middle ear exudate is well established[8,13]. But not many researches are available on co-relation between tonsillitis and middle ear exudate in cases of chronically discharging ears. Thus, this study was done to explore the relationship between bacterial flora of tonsil and a discharging ear.

Material and methods:
About 157 Patients with ear discharge attending Index medical college and hospital were taken into study during the period of one year from April 2022 till April 2023 and under asepsis swab was collected from the throat as well as discharging ear. Those with dry ear or having rhinosinusitis were excluded from study. About 1080 (17.15%) cases out of 95631 patients attending ENT OPD had discharging ear (both acute and chronic Suppurative Otitis Media). Age of patient, or any other co-morbidity was not an exclusion criteria. The patient having throat infection or even normal throat were also included in the study irrespective of acute or acute on chronic infection in throat.
Culture swabs were taken from the throat (posterior pharyngeal wall) ensuring that while taking swab, it doesn't touch buccal mucosa. For ear swab, any excess discharge was removed from the external auditory canal and EAC was cleaned with sterile cotton pledget soaked in alcohol. Swab sample was taken using sterile albumin coated tip on an applicator from bony portion of EAC.
These swabs were transported in sterile tube with Amies Transport Media for transportation to Department of Microbiology for culture and gram stain study.
Gram staining of all the smears prepared from these swabs was done and bacteriological culture was done over blood agar, MacConkey agar and Chocolate agar media plates. Innoculation of material was done using Bichrome Loop or Platinum loop. These culture plates were incubated at 37^0 C for 18 - 24 hours and growth of colonies noted. Those sterile cultures were reincubated for another 24 hours to see if any growth develops. Chocolate agar plates were kept in candle jar to provide microaerophilic environment required for H. influenzae growth.
Colonies were identified, smears were made and further processing of biochemical test battery comprising of Catalase test, Oxidase Test, Coagulase test etc.

Results:
Out of 1080 cases with ear discharge, only 157 cases having ear discharge were taken in study. Of these 53 cases (33.75%) were having complaint of sore throat whether tonsillar infection or tonsillitis and remaining 104 (66.24%) cases had a normal throat.
Clinically, none of the case had Acute Suppurative Otitis Media, though 26 (16.67%) cases had Acute on Chronic Suppurative Otitis Media with pulsatile serosanguinous discharge. Sixty-two (39.74%) cases has copious, nonfoul smelling, thick Mucopurulent discharge and also had central perforation. While 68 (43.59%) had scanty foul-smelling discharge with either retraction pocket or attic perforation suggestive of squamosal or unsafe type of suppurative otitis media.
This study had male dominance with 89 (56.69%) and females 68 (43.31%). Most of the cases were between 21-30 years (52 cases/ 33.12%). Followed by 11-20 yrs (30.57%), 31- 40 yrs. (16.56%) and least in 41 years and above 8.28%. Youngest patient was of 3 years and oldest being 45 years. Maximum males were in age group of 21-30 years (19.11%) and females were in age group of 11-20 years (38.23%). (Table I).

Age Group	No of cases (%)	Males (%)	Females (%)
00-10 years	18 (11.46)	14 (8.9%)	4 (2.56%)
11-20 years	48 (30.57)	22 (14.02)	26 (16.56)
21-30 Years	52 (33.12)	30 (19.11)	22 (14.01)
31-40 Years	26 (16.56)	15 (9.51)	11 (7.01)
40 Years & above	13 (8.28)	8 (5.1)	5 (3.18)
Total	157 (100)	89 (56.69)	68 (43.37)

Table I : Table showing age and sex distribution of cases

Culture swabs were collected from throat and ears of all the cases and culture growth with biochemical testing was done. Smears were made from all the swabs and Gram's staining was done. Gram's staining of smears prepared from throat sweab showed presence of Gram-Positive Cocci clusters in 153 (97.45%). Gram positive chain in 146 (92.99%), Gram Positive Cocci (Large) in 11 (7%), Gram Positive in pairs in 38 (24.2%) and Gram-Positive Bacillus in 24 (15.3%). While Gram Negative Bacilli was seen in 45(28.66%) smears with Gram Negative Cocci Bacilli in 6(3.82%) and Gram-Negative Cocci in 15(9.5%). Ear swab Gram staining showed Gram Positive Cocci clusters in 79 (50.3%). Gram positive chain in 6 (3.82%), Gram Positive Cocci (Large) in 1 (0.63%), Gram Positive Bacilli in 16 (10.19%). While Gram Negative Bacilli was seen in 110 (70.06%) smears and Gram- Negative Coccobacilli in 14 (8.91%) Table 2).

Gram Stain	Throat Swab (%)	Ear Swab (%)
Gram Positive Cocci		
1. Cluster	155 (98.73)	79 (48.59)
2. Chain	146 (92.99)	06 (3.82)
3. Large	11 (7.0)	01 (0.63)
4. Pairs	38 (24.2)	00
5. Bacilli	24 (15.3)	16 (10.19)
Gram Negative		
1. Bacilli	53 (33.76)	110 (70.06)
2. Cocci	18 (11.46)	00
3. Cocobacilli	9 (5.73)	14 (8.91)

Table II : Table showing Gram staining of swabs from throat and ears

Inoculation growth of throat swab after 24 hours on culture plate showed Gram Positive Cocci clusters in 151, Gram positive chain in 144, and Gram-Positive Bacillus in 24 (15.3%) cases, while Gram Negative Bacilli grew in 44 (28.66%) smears with Gram Negative Cocci Bacilli in 6(3.82%). Similarly, colony smear from ear swab grew Gram Positive Cocci clusters in 58 (37.57%). Gram positive chain in 4 (2.54%), Gram Positive Cocci (Large) in 1 (.063%), Gram Positive in pairs in 14 (8.91%) and Gram-Negative Bacilli in 101 (65.60%) smears with Gram Negative Cocci Bacilli in 6(3.82%) and Gram-Negative Cocci in 15(9.5%) (Table III).

Gram Stain	Throat Swab (%)	Ear Swab (%)
Gram Positive Cocci		
1. Cluster	151 (97.45)	58 (37.57)
2. Chain	144 (92.99)	04 (2.54)
3. Large	11 (7.0)	01 (0.63)
4. Pairs	38 (24.2)	00
5. Bacilli	24 (15.3)	14 (8.91)
Gram Negative		
1. Bacilli	44 (28.66)	101 (65.60)
2. Cocci	15 (9.55)	00
3. Cocobacilli	6 (3.82)	12 (7.64)

Table III : Table showing Gram staining of Cultures from throat and ears

Throat swab culture grew minimum 2 organism in 65 (41.40%), three in 77 (49.05%) and four organisms in 15 (9.55%) cases, while in culture from ear swabs single organism was grown in 121

(77.01%), two in 15 (9.55%) and three in 13 (8.28%) cases.

On identification of bacterial using biochemical reactions and colony characteristics, throat swab revealed maximum growth with Strepto. viridance in 126 (86.25%), followed by Staph. Aureus 94 (59.87%). Eriwinia grew in 1 (0.63%) case. Out of 52 swabs from infected throat, Staph aureus grew in 51 cases and24 had Staph. epidermidis and only 1 swab Eriwinia and Acineto antitrtus (table IV). From 105 normal throat cases, Strepto. viridans grew in all the 105 cases, followed by Staph. aureus in 43 swabs.

Organism cultured	Infected throat (n=52)	Normal throat (n=105)
1. Staph. aurues	51 (54.26)	43 (45.76
2. Staph. epidermidis	24	15
3. Strepto. Viridans	21	105
4. Strepto. Pyogenes	16	4
5. Micrococcus	3	4
6. Pneumococcus	13	8
7. Diphtheroids	7	17
8. Kleibsella aerogenes	7	26
9. Ps. Aeruginosa	2	4
10. E. coli	2	3
11. Erwinia	1	0
12. Acineto anitratus	1	5
13. B. catrrhalis	4	11

Table IV : Table showing organisms grown from swabs from throat and ears

Ear swab culture identifies Staph. aureus in maximum cases in 59 (37.57%) cases followed by Pseudomonas aeruginosa in 53 (33.76%) cases. Uncommon bacterias like Micrococus, Citerobacter freutidis were also grown in 1 case each. Sterile culture even after incubation of 72 hours was obtained in 7 cases (Table V).

Of the 27 acute on chronic and mucosal suppurative otitis media Staph. aureus grew in maximum 16 and 36 cases respectively, while in squamosal Suppurative otitis media it was only in 7 out of 68 ears swab. In squamosal type of disease Pseudomonas aeruginosa grew in maximum 34 cases of 68 cases, and in mucosal disease it grew in 19 (3 in acute on chronic and 16 on chronic mucosal disease).

E.coli grew in only 3 of acute on chronic cases and none in chronic or squamosal type of SOM, similarly Citerobacter grew in only one case of squamosal disease and Acinetor lwoffi in one case of each type of ear discharge.

Organism cultured	Total (N=157)	Ac on Chronic (n=27)	Mucosal (n=62)	Squamosal (n=68)
4. Staph. aurues	59	16	36	7
5. Strepto. Pyogenes	04	01	03	00
6. Kl. aerogenes	19	00	14	07
7. Ps. Aeruginosa	53	3	16	34
8. Micrococcus	01	00	01	00
9. E. Coli	01	03	00	00
10. Diphtheroids	14	1	3	10
11. Proteus vulgaris	14	3	11	00
12. Proteus mirabilis	12	1	11	00
13. E. coli	3	3	00	00
14. Citerobacter	01	00	00	01
15. Acineto lwoffi	03	01	01	01
16. Sterile	7	1	5	1

Table V : Table showing organisms grown from ear swab with types of ear disease

Statistically checking co-relationship between throat and microbial flora & ear swab 87 (55.41%) showed presence of similar organism in both ear and throat of which 41 (78.841%) cases had throat infection and 46 (45.09%) had normal throat. Similar organisms were cultured in mostly safe type of disease with 55 ears having safe or mucosal type of suppurative otitis media, 21 having acute on chronic safe or mucosal type of suppurative otitis media. While only 11 (16.17%) cases with similar bacterial flora had unsafe or squamosal chronic suppurative type of infection.

There are no direct literature available on co-relation between throat and ear discharge bacterial flora. Though Maw & Speller 1985[12] tried to establish relationship between glue ear and infection of tonsils and adenoids. They established that there is not much difference in bacterial flora of adenoid & tonsills with or without infection and they could not find nay stastically significanty relationship between two. However, in our study using Chi square (X^2) correlation between throat and ear discharge bacterial flora we found significant relationship (X^2=17.2, DF=1) P>0.05 and similar significance was found in infected throat and ear discharge (~~Z~~=5.10, P>0.05). While statistical significant relationship was absent between normal throat and discharging ear (~~Z~~=1.28, P<0.01). while calculating statistically significance betwen various type of suppurative otitis media and throat swab; a significant relationship was present between Acute on chronic mucosal suppurative otitis media and bacterial flora of throat X^2=4.29; DF=1 and P>0.05 with yates correction. Similar significance was seen between mucosal chronic suppurative otitis media with X^2=14.3, DF=1 with P>0.01. However, in unsafe or squamosal type of chronic suppurative otitis media dis not showed any statistically significant relationship with bacterial flora of throat irrespective of presence or absence of throat infection (X^2=2.81, DF+1 & P<0.01).

Discussion:

Through there is a well-established relationship between organisms of nasopharynx and ear infection, but there is no direct co-relationship between throat and ear infection were studied and nor many literatures are available. We tried to find the bacterial relationship between throat bacterial flora and organisms present in ear discharge, to establish that even infection from throat can be a cause of ear infection.

Of the 157 cases we studied, we found that 3rd decade has maximum 52 cases (33.12%) followed by 2nd decade 48 (30.75%) and combined 63.87%. This was similar to study by Rao & Reddy (1994)[3] who also reported 69.17% maximum cases in first three decades combine. Shyamal R (2012)[8] & L. Appiah-Korang et al (2014)[15] also reported combined 70.96% cases in 1st two decades of life, similarly Alem Getaneh et al (2021)[9] and Monasta L (2012)[2] in their study of CSOM also reported higher incidence in first 3 decades of life. Though Constable & Butler (1982)[16] & Xu J (2021)[17] reported higher incidence in 5th and 6th decade of life.

Though all the studies and our studies reported higher incidence in
Males[3,7,14,15,16,17,18], except Khatun MR et al (2021)[19] reported higher incidence of female in their study with female dominance in ratio of 1.2:1 to males.

A positive culture was seen in 95.54% ear swabs in our study which was also seen in study of Prakash R (2013)[18] 91.8% and Shyamal R (2012)[8] 93%, this was higher than the other studies of L-Appiah korang (2014)[15], Xa ju (2021)[17]and Khatun MR (2021)[18], where they found more sterile cultures ranging from 23-30% plates. In our study single organism growth was seen in 77.07% ear swabs which was similar to finding of Rao & Reddy (1994)[3] Xu J(2021)[17], L. Appiah Korang et al (2014)[15] A. Getaneh (2021)[8] all reported higher incidence of infection with single organism with range of 91-95%. This was much higher than our study. Staphylococcus aureus (37.5%), Pseudomonas aeruginosa (33.75%) followed by Klebsiella aerogenes (12.75%) and Diptheroids (8.91%) were the main organisms grown from ear discharge swab culture. Almost all the authors like Rao & Reddy (1994)[3] , Xu J et al(1921)[17], L. Appiah Korang et al (2014)[15] Madana J et al (2011)[20],

A. Getaneh et al (2021)[9] reported similar pattern of growth in their studies. However, there was minor variation in the frequency of growth of Proteus and Diptheroids in some studies.

Throat swabs culture showed Streptococcus viridance followed by Staph. aureus was the most cultured bacteria which was also reported by Chaturvedi et al (1985)[21], Maw & Speller (1985)[22], Rusan M Klug (2009)[23] and Jitendera Yadav et al (2015)[24] . While Jose M Odonezo Mera etal (2020)[25] found Hemophilus influenzae followed by Streptococcus pneumonae as most common bacteria grown from throat swab.

Since there were no literature available on establishing relation between bacterial flora of throat and organism cultured from ear swab. Though Maw & Speller (1985)[22] had done a study on Glue ear and established that bacterial flora of tonsils and adenoids are same. In our study we found that of 87 (55.41%) cases of which 41 (45.98%) cases were having throat infection and 46 (54.02%) were having normal throat have same bacterial flora as of ear swabs. Statistically, we found a significant relationship (X^2 =17.2, DF=1) between throat infection and ear discharge (~~Z~~=5.10, P>0.05). While no statistical relationship was present in normal throat and ear discharge (~~Z~~=1.28 , P<0.01). Similarly no statistical relationshship was found between unsafe or squamosal chronic suppurative otitis media and throat was (X^2 =2.8, DF=1) with P>0.01.

Thus, we conclude that there is a significant chance that the organism in the throat especially during acute throat infection can be held responsible for the infection in middle ear causing mucosal or safe type of suppurative otitis media

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