Comparing Cerumen Bacterial Flora in Acute Otitis Externa Patients and Healthy Controls

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Abstract

Introduction:
Cerumen is known as ear wax, produced regularly by cerumen and lipid secretory glands. Regarding the effect of Mazandaran province's humid weather on the prevalence of pathogenic microorganisms, this study was performed to determine the bacterial flora of the ear in patients with acute otitis externa and its comparison with healthy subjects.

Materials and Methods:
In this case-control study, cerumen was collected and cultured from 40 patients with clinically diagnosed acute otitis externa and 80 healthy controls. The data were finally analyzed using SPSS.

Results:
In the study group, Staphylococcus aureus (20.8%), Bacillus (18.9%) and Pseudomonas (11.3%) and in the control group Staphylococcus epidermidis (38.7%) and Diphtheroid (22.4%) were the most common bacteria, respectively.

Conclusion:
The isolated bacteria from cerumen of healthy subjects were totally different from those of acute otitis externa patients.

Keywords:
Bacteri, Cerumen, Otitis extern

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Introduction
Cerumen or ear wax is secreted by ceruminous and sebaceous glands. Two types of human cerumen, wet and dry, are controlled by two autosomal alleles (1). However, some differences have been observed between cerumen components such as amino acids, cholesterol, triglyceride, lysosome based elements, immunoglobulin, glycopeptides, copper and other components which are the constituents of cerumen (1). The cerumen function in protecting the ear against microorganisms has been an issue of discussion for a long time. On one hand, it is said that cerumen is not able to prevent infection while rich nutrients of the ear wax cause an increase in bacterial and fungal quantity. On the other hand, it is said that cerumen may have antibacterial activities. Nevertheless, there is little evidence to support this concept in either way (2).

We aimed this study on comparing the normal bacterial flora of cerumen with that of acute external otitis patients in Mazandaran province.

Materials and Methods
This study was conducted as case-controls. Acute otitis externa cases which were referred to the ear, nose and throat (ENT) unit of Shahid Beheshti Hospital, Babul, Iran, were enrolled in the study. Healthy cases without any medical problems but with cerumen in the ear canal were recruited as the control group. After excluding ineligible cases, 40 patients and 80 controls entered the study protocol and filled an informed consent. Clinical examination by the aid of a qualified microscope was performed and the patient's medical history was fully reviewed by an ENT specialist. Cerumen samples were obtained from all cases by suction, curettage or the use of a loop. These samples were then sent to the microbiology department of Babul University of Medical Sciences in sterile transfer medium where the samples were cultured on chocolate agar, blood agar and eosin methyl blue (EMB) media. The chocolate agar and EMB agar were incubated for 18-24 hrs at 37°C under Co2 condition. Colonies characteristics were then examined by direct microscopic examination (gram staining) and also special diagnostic tests.

Statistical analysis: Data were inserted into SPSS software Version 16 and analyzed by using Chi-square and t-Tests. A P-value equal to or less than 0.05 was considered as significant.

Results
The mean age was 39.53 and 36.28 years for patients and healthy controls, respectively. No significant difference in age existed between the two groups (P=0.136).

The study and control group consisted of 11(27.5%) and 25(31.3%) males and 29 (72.5%) and 55(68.7%) females, respectively. The two groups showed no significant difference based on age (P=0.42).

The frequency of monobacterial and poly bacterial flora in the study and control groups is listed in (Table 1).

98.3% and 1.7% of bacteria were gram positive and gram negative in the control group whereas these rates were 79.2% and 20.8% in the study group, respectively (P=0.000).

Table1: Monobacterial, polybacterial and nonbacteria in the healthy and patient group

<table>
<thead>
<tr>
<th></th>
<th>Patient group number (percentage)</th>
<th>Healthy group number (percentage)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monobacterial</td>
<td>24 (60)</td>
<td>48 (60)</td>
</tr>
<tr>
<td>Polybacterial</td>
<td>14 (35)</td>
<td>29 (39.25)</td>
</tr>
<tr>
<td>Nonbacterial</td>
<td>2 (5)</td>
<td>3 (3.75)</td>
</tr>
</tbody>
</table>

Staphylococcus aureus, Klebsiella pneumonia, Streptococcus pneumonia, Haemophilus influenzae and Pseudomonas are considered as pathogenic bacteria of the external ear, other bacteria are regarded as the normal flora. Accordingly, 39.6% of bacteria in the study group were pathogenic while 60.4% were of normal flora; in the
study group 6.9% of the bacteria were pathogenic and 93.1% were from normal flora (Table 2).
The most prevalent bacteria in the two groups are listed in Table 3. The prevalence of pathogenic bacteria in the study and control groups were 39.6% and 6.9%, respectively ($P=0.000$).

### Table 2: Pathogen and normal flora bacteria in the healthy and patient group

<table>
<thead>
<tr>
<th></th>
<th>Healthy group</th>
<th>Patient group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacterial flora</td>
<td>108 (93.1%)</td>
<td>8 (6.9%)</td>
</tr>
<tr>
<td>Pathogen</td>
<td>8 (6.9%)</td>
<td>21 (39.6%)</td>
</tr>
<tr>
<td>Total</td>
<td>116 (100%)</td>
<td>32 (60.4%)</td>
</tr>
</tbody>
</table>

The number of bacterial colonies based on the type of bacteria in both groups is shown in Table 4. As is seen in the study group, the colony count in 13 cases (24.6%) was less than 10 CFU whereas in 40 other cases (75.4%) it was numerous. *Staphylococcus aureus*, *Bacillus*, and *Pseudomonas* were the most common bacteria with a high colony count.

In the control group, colony count was less than 10 CFU in 37 cases (31.9%), 10-100 CFU in 16 cases (13.8%) and numerous in 63 cases (54.3%). *Staphylococcus epidermidis* and *Diphtheroid* were the most common bacteria with a high colony count and of a nonpathogenic type in the ear.

### Discussion

In this study, monobacterial, polybacterial and nonbacterial isolates in the control group were 60%, 36.25% and 3.75%, respectively (Table 1); while in the study group the same isolates were 60%, 35% and 5%, respectively. Similar to our study, in Pata's study (3), monobacterial and polybacterial isolates in the control group were 65% and 35%, respectively. In Clark et al study (5), monobacterial and polybacterial flora of the patients were 57% and 43%, respectively. Therefore, our results conform to such similar studies to a high extent. However, in Stroman's study (4), monobacterial isolates were seen in 33.4% of cases while polybacterial isolates were detected in 66.6% which does not conform to our results. This difference could be due to the ear anatomy, the geographic area of residence or the ability of an individual or laboratory techniques in isolation and diagnosis of bacteria.

The absence of bacterial growth in two groups of some samples both in our study and in other studies (3) may be due to a viral infection such as *Herpes simplex virus* and *Papovavirus*, existence of anaerobic bacteria or various diagnostic approaches (6).

In the current study, the most common bacteria in the patient group were *Staphylococcus aureus* (20.8%), *Bacillus* (18.9%) and *Pseudomonas* (11.3%). While *Staphylococcus epidermidis* (38.7%), *Diphtheroid* (22.4%) and *Streptococcus* (22.4%) were the most common types of bacteria in the control group. These results conform to those of Dibb (7), Stroman, Campos (4,8) and Pata’s studies (3).

*Pseudomonas* species and *Staphylococcus aureus* were the two common bacteria isolated from the study group which also conform to Loh’s study (9). Among the 26 patients in Clark et al study, *Pseudomonas* was observed in 14 cases (53.8%), *Staphylococcus* in 7 cases (26.9%), *Acinetobacter* and *Enterococcus* both in 2 cases (7.6%) (8).

Also, there is conformity concerning *Staphylococcus aureus* and *Acinetobacter*. In contrast to Clark’s study, *Pseudomonas* was isolated from 11.3% cases. This can be due to variety in the type of pathogen in different geographical areas; in our study the isolated pathogens were *Streptococcus pneumoniae*, *Klebsiella pneumonia* and *Haemophilus influenzae*, whereas such pathogens were not isolated in Clark’s study (5).

*Staphylococcus aureus* is one of the pathogenic bacteria which was isolated from 11 cases (20.8%) in the study group and in 8 cases (6.9%) of the control group; it was also detected in healthy participants in Stroman and Campos’ study (4,8). It is worth mentioning that in the 8 cases of the control group in which *Staphylococcus aureus* was
observed the colony count was less than 10 CFU in all 8 cases according to table 5 which is negligible from the point of bacteriology. Moreover in 10 of 11 cases in the patients' group *Staphylococcus aureus* showed an abounding number of bacteria. The number of colonies in the patients' group was less than 10 CFU in 24.6% of cases and abounding in 75.4% of cases. It means that most isolates from the cerumen of patients had a high colony count. This quantity was considerable for *Staphylococcus aureus* (91% of cases with abounding colony) and pseudomonas (83.3% of cases with abounding colony). In comparison between the two groups, the number of colonies had a wider range in the control group; in most cases it was less than 10 CFU, 10-100 CFU or numerous colonies from one type of bacteria, which shows a normal floral state.

Overall, the results of our research conform to similar studies in most cases and small differences could be attributed to difference in geographical and residential areas, race, living status and socio-cultural position and also to the various sampling and diagnostic approaches. Also, it seems that viruses should be considered as probable pathogens in patients with otitis externa.

**Conclusion**

The isolated bacteria from cerumen of healthy subjects were totally different in comparison to those of acute otitis externa patients.

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**References**