

STUDIES ON THE GROWTH OF BACTERIA IN THE HUMAN EAR CANAL*

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The relationship between bacteria and external otitis in man is vague. Occasionally bacteria are directly and completely responsible for the disorder and antibacterial therapy, as indicated by culture and sensitivity tests, will effect an immediate and satisfactory recovery. Sometimes bacteria play only a secondary role in the production or protraction of external otitis, e.g., although pathogenic organisms are cultured, antibacterial therapy produces only a partial response. Usually, even though microorganisms can be cultured from the external auditory canal, they are found to be completely unrelated to the disease process.

The mere presence of microorganisms on culture is not tantamount to a diagnosis of bacterial external otitis. It is considerably easier to culture the bacteria than it is to assign an etiologic role to them with any degree of certainty. A knowledge of the bacterial flora of the human ear canal in health is essential in evaluating the possible etiologic significance of organisms cultured from the diseased canal. The first portion of this study is a survey of the aerobic and anaerobic flora of the healthy human ear canal.

A second focus of interest in considering the role played by bacteria in the production of external otitis centers around the possibility that cerumen may possess a bacteriostatic property. We have investigated this possibility by using a method commonly employed to determine the sensitivity of bacteria to antibiotics.

The literature is replete with references to the production of external otitis by *Pseudomonas aeruginosa*. A number of authors (1, 2, 3) feel that this organism is responsible for a large percentage of clinical otitis externa. To investigate this, experimentally virulent organisms of *Pseudomonas aeruginosa* were seeded into the ear canals of healthy volunteers.

METHODS AND MATERIALS

Bacterial flora: Cultures were taken from both external auditory canals of forty-five healthy adult volunteers: twenty-five hospital employees and twenty inmates of a penal institution. The culture swabs were immediately placed in test tubes containing 0.5 cubic centimeter of brain heart infusion broth and cultures were set up within four hours. Gram stains were made of the broth inoculum. Direct cultures of one loopful of the inoculum were made in duplicate to blood (horse) agar. Brain heart infusion broth was inoculated in duplicate with two-tenths cubic centimeter of the broth inoculum. The cultures were incubated under aerobic and anaerobic conditions. The Brewer method of anaerobiosis was used.

After growth for twenty-four hours, the aerobic cultures were read. All questionable colonies were stained by Gram method and subcultured to broth. If the aerobic broth

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cultures showed gram negative rods, Eosin Methelene blue and S. S. agar were also inoculated.

The anaerobic cultures were grown for forty-eight hours before reading. Subcultures of the broth culture were made to blood agar and incubated under anaerobic conditions for another forty-eight hours. Biochemical studies were made of five of the seven coliform organisms. Coagulase tests were made of all *Micrococcus aureus* and many of the *Micrococcus albus* organisms.

Cerumen: Cerumen was collected from the ear canals of twenty healthy adult subjects. Random samples of the pooled specimen were cultured in broth and on blood agar plates. Other portions were streaked in a band across one diameter of each of several blood agar plates. Organisms were then streaked across these plates in parallel lines perpendicular to and crossing the band of cerumen. Because of their frequent occurrence in healthy or diseased ear canals, the following bacteria were chosen:

1. *Micrococcus aureus* (hemolytic)
2. *Streptococcus pyogenes*
3. *Corynebacterium*
4. *Micrococcus albus* (hemolytic)
5. *Bacillus subtilus*
6. *Escherichia coli*
7. *Pseudomonas aeruginosa*

Pseudomonas aeruginosa: Organisms of a virulent strain of *Pseudomonas aeruginosa* were liberally seeded into the external auditory canals of seven healthy volunteers who were instructed to keep water out of their ears and to keep their hands away from them. After one week, cultures from these canals were examined by the same methods used to study the flora of the healthy canal.

RESULTS

Bacterial flora: The incidence of the organisms making up the flora of the human external auditory canal in health is shown in Table I. It will be noted that

TABLE I
Bacterial flora of ninety normal external auditory canals

Organism	Incidence
Hemolytic <i>Micrococcus albus</i>	78
<i>Micrococcus albus</i>	66
Hemolytic <i>Micrococcus aureus</i>	11
<i>Micrococcus aureus</i>	6
<i>Micrococcus citreus</i>	1
<i>Streptococcus alpha</i>	1
<i>Streptococcus beta</i>	1
<i>Streptococcus gamma</i>	2
<i>Corynebacterium</i> (hemolytic).....	21
<i>Corynebacterium</i> (non-hemolytic).....	73
Coliform.....	7
<i>Proteus</i>	1
<i>Clostridium</i> (hemolytic).....	1
<i>Sarcina</i>	3
<i>Bacillus</i>	14
<i>Gaffkya</i>	1
Unidentified.....	1

* The total incidence does not correspond to the sum of individual incidences because many canals yielded more than one organism in each group.

the *Micrococcus* was found in every ear canal. The majority of the examples of *Micrococcus aureus* were coagulase positive as was an occasional *Micrococcus albus* (hemolytic).

Six of the ear canals cultured yielded a pure culture of a single organism. Sixty canals gave cultures of two organisms only and twenty-four grew out three or more different organisms. Of the seven coliform organisms, five were identified as aerobacter. The one unidentified organism was a short gram negative rod which grew on blood agar as a small, transparent colony resembling streptococcus.

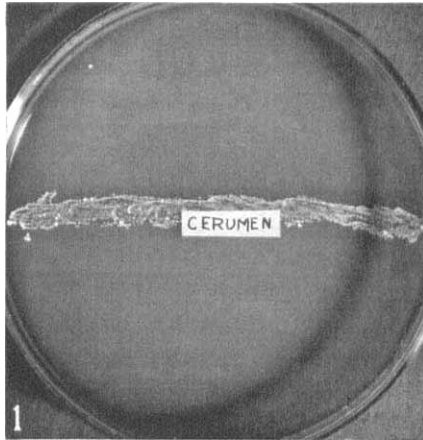
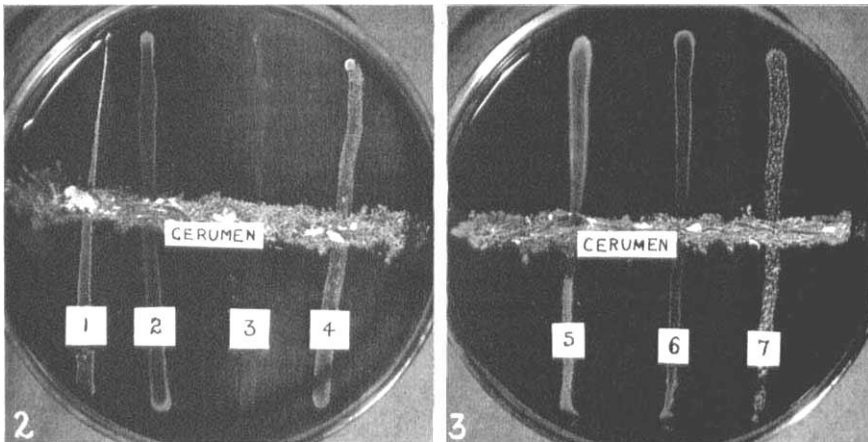


FIG. 1. A band of cerumen streaked across a blood agar plate. The cerumen represents a pooled specimen from twenty subjects. The colonies of bacteria growing from the cerumen are *Micrococcus albus* and *Corynebacterium*.



FIGS. 2 and 3. Two blood agar plates with a band of cerumen running across each plate. The numbered lines are cultures of organisms representative of the flora of healthy and diseased ear canals: 1) *Micrococcus aureus* (hemolytic), 2) *Streptococcus pyogenes*, 3) *Corynebacterium*, 4) *Micrococcus albus* (hemolytic), 5) *Bacillus subtilis*, 6) *Escherichia coli* and 7) *Pseudomonas aeruginosa*. There is no evidence of inhibition of bacterial growth by the cerumen.

Since the difference between the cultures of hospital employees and of the inmates of the correctional institution were minimal, the results have been combined in the tabulation.

Cerumen: From the blood agar plate (Figure 1) and from the broth cultures of cerumen, the following three organisms were identified: 1) *Micrococcus albus* (hemolytic), 2) *Micrococcus albus* (non-hemolytic) and 3) *Corynebacterium*.

Figures 2 and 3 demonstrate that there was no inhibition of growth of any of the microorganisms that were cultured on blood agar plates crossed by the band of cerumen.

Pseudomonas aeruginosa: One week after seeding *Pseudomonas aeruginosa* into the ear canals of the subjects culture of the canals failed to reveal the presence of the organism in any of the seven subjects. There was no clinical evidence of external otitis.

DISCUSSION

Bacterial flora: With few exceptions, our findings are in general agreement with those of other workers who have studied the bacterial flora of the human external auditory canal in health. The particularly good general agreement with the survey of Singer and his coworkers (1) is of interest because it affords an opportunity to compare the flora in a temperate climate (Philadelphia) with that found in a sub-tropical climate (Florida). The organisms that we found are, in general, the same ones found by Haley (4) except that the incidence of each organism is higher in the present series than she reported in her survey. There is also good correlation with the results published by Syverton *et al* (5) except that after examining only sixteen cases, they did not report many of the more infrequently occurring organisms.

At the outset it was felt that differences in personal hygiene and in living standards might dictate differences in the flora of hospital employees and inmates of the penal institution. This was not borne out in the results. The only differences seen between the two groups were in the decreased frequency of some of the transients in the ear canals of the hospital employees. The resident flora is probably identical in the two groups.

While only one obligate anaerobe was found (a *Clostridium*, hemolytic, grown from one ear), the anaerobic methods contributed to the information gained inasmuch as they increased the incidence of many organisms. Furthermore, anaerobic cultures are desirable if not mandatory in studying the diseased ear canal. As previously stated elsewhere (6), anaerobic cultures are indicated in studying the bacterial flora of any area of the skin if pathogenic organisms are to be demonstrated in the highest possible incidence.

The low incidence of Streptococci in this series and in Singer's survey is notable. These organisms are rarely found on normal skin and their presence in the face of a dermatitis of the ear canal must therefore be considered to be significant. This is particularly true of beta-hemolytic Streptococci.

There were no organisms in the flora of the ear canal that have not been found as either resident or transient organisms on the skin elsewhere (7).

In the present series there were sixty-eight male subjects and twenty-two

female. The resident flora did not differ between the sexes. There was a minor variation in that the male subjects tended to carry a slightly higher incidence of transient organisms in their canals.

Although the survey covered a period of nine months (from August to April) there was no evidence of seasonal variation in either the resident or transient bacterial population of the ear canal.

Cerumen: The isolation of *Micrococcus albus* and *Corynebacterium* from cerumen is consonant with the findings of Creed and Negus (8) although they also found sarcina and gram negative diplococci in a small percentage of their subjects.

A number of clinicians who have a rich experience in the treatment of external otitis have noted a paucity or absence of cerumen in the ear canals of patients with the disorder. They deduced that the increased numbers and strains of organisms cultured from diseased canals were a result of the absence of cerumen and ergo, that cerumen must possess some bacteriostatic property. They thought the failure of sebaceous and ceruminous glands to produce cerumen was one of the initial pathologic mechanisms in the development of bacterial external otitis.

Conley (9) has published a review of the work of a number of investigators who have helped to dispel this erroneous concept. Creed and Negus (8) felt that cerumen had no bactericidal action. Pirodda (10) stated that it was effective against pneumococci and diphtheria bacilli only. We used five organisms frequently found in the healthy ear canal and two organisms that are often isolated from diseased canals (hemolytic *Streptococcus* and *Pseudomonas aeruginosa*) to test the theory. There was no evidence of inhibition of the growth of any of these organisms.

Pseudomonas aeruginosa: Hardy and his co-workers (11) commented on the rarity of *Pseudomonas aeruginosa* in the normal external auditory canal when compared with its occurrence in large numbers in a high percentage of exudative cases of otitis externa. They concluded that this implicated the *Pseudomonas* as the sole or associated cause of a very high proportion of their cases of external otitis.

Salvin (12) reported that he was unable to produce an external otitis in the ears of young albino rabbits with a saline suspension of *Pseudomonas aeruginosa* unless he had previously traumatized the skin of the ear. He used organisms originally isolated from the ears of patients with external otitis.

The absence of clinical external otitis and our inability to culture *Pseudomonas aeruginosa* from the ear canals of volunteers seven days after they had been liberally applied to the skin is consonant with the findings of Salvin. It leads us to the conclusion that the mere presence of *Pseudomonas* on the skin of the ear canal is not sufficient cause for a dermatitis of the canal wall. This bears a further implication in the logical management of external otitis believed to be due to *Pseudomonas*. Antibacterial measures alone are not likely to be adequate treatment. All trauma to the skin of the canal must be prevented if the skin is to recover and to remain well. The patient must not be subjected to the added stress of high environmental heat or humidity.

CONCLUSIONS

1. The resident bacterial flora in the healthy human ear canal is remarkably constant. There is no difference in the flora in various geographic locations, between the sexes or from season to season. The resident flora is composed primarily of Micrococci and Corynebacteria. The transient flora varies slightly with personal hygiene.

2. Cerumen, as it exists in the ear canal in health, does not, by the method used, exhibit any inhibition of the growth of the organisms tested.

3. *Pseudomonas aeruginosa*, acting alone, is not likely to be the cause of external otitis in previously healthy ear canals. If it is an etiologic factor, secondary etiologic agents such as trauma or increased environmental heat or humidity, must be required to act simultaneously.

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