Pathologic Findings of the Cochlea in Labyrinthitis Ossificans Associated with the Round Window Membrane

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Abstract

Objective. To quantitatively demonstrate and classify the histopathologic changes in the cochlea of the human temporal bones with labyrinthitis ossificans (LO).

Study Design. Comparative human temporal bone study.

Setting. Tertiary academic medical center.

Subjects and Methods. We compared 23 temporal bone specimens from 19 deceased donors with LO associated with the round window membrane (RWM) and 27 age-matched specimens from 20 deceased donors without any otologic diseases. We focused on the location of LO in the inner ear, the intensity of endolymphatic hydrops, the number of spiral ganglion cells and cochlear hair cells, and the areas of the stria vascularis and spiral ligament. In addition, we created a new pathologic grading system for temporal bone specimens from deceased donors with LO associated with the RWM.

Results. We most often observed LO in the scala tympani of the basal cochlear turn. In the LO group (as compared with the control group), the intensity of endolymphatic hydrops was significantly increased; the number of spiral ganglion cells was significantly decreased in all segments; the loss of outer and inner hair cells was significantly increased in all turns of the cochlea; the atrophy of the stria vascularis was significantly greater in all turns of the cochlea; and atrophy of the spiral ligament was significantly greater in the basal and middle cochlear turn.

Conclusion. LO was associated with significant cochlear damage (to the spiral ganglion cells, cochlear hair cells, stria vascularis, and spiral ligament) and with increased intensity of endolymphatic hydrops.

Keywords

labyrinthitis ossificans, round window membrane, cochlea, endolymphatic hydrops, spiral ganglion cell, outer hair cell, inner hair cell, stria vascularis, spiral ligament, cochlear implant, human temporal bone, histopathology

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Labyrinthitis ossificans (LO) refers to pathologic ossification within the lumen of the otic capsule, including the cochlea and vestibule, caused by an inflammatory or destructive process. Paparella and Sugiura divided the progression of pathologic new bone formation into 3 stages: acute, fibrous, and ossification.1 Ossification starts to develop in the perilymph of the basal turn and finally can involve the whole inner ear. In animals, ossification can be seen 2 months after the acute stage; however, in humans, it takes a long time.1

Trauma and several diseases have been associated with LO, such as tumors, intralabyrinthine hemorrhage, otosclerosis, osteomyelitis, sickle cell disease, and autoimmune inner ear diseases.2-4 The most common cause of LO is infection, which can come via 3 routes: tympanic, hematogenous, or meningeal.5 LO might be an end-stage sequel of suppurative labyrinthitis and can lead to sensorineural hearing loss and vestibular dysfunction.2

One of the options for treating patients with profound sensorineural hearing loss is a cochlear implant. Because a cochlear implant involves the insertion of the electrode through the round window membrane (RWM), any ossification or blockage at the scala tympani can affect the course of the surgery and its results. In this study, our primary aim was to obtain quantitative data on the status of cochlear structures in temporal bone specimens from deceased donors with LO associated with the RWM—with the ultimate goal of improving the evaluation of cochlear implant

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candidates with known LO. As part of this study, we described the characteristics of LO in temporal bone specimens with the RWM ossification and created a new pathologic grading system.

**Materials and Methods**

We obtained specimens for this study from the archived human temporal bone collection at the University of Minnesota. The temporal bones that we used were removed at autopsy and fixed in formalin solution. Each bone was decalcified, embedded in celloidin, and then serially sectioned in the horizontal plane at a thickness of 20 μm. Every 10th section was stained with hematoxylin and eosin and mounted on a glass slide for light microscopic observation. This study was approved by the Institutional Review Board of the University of Minnesota (0206M26181).

In this study, we compared 2 groups: the LO group (comprising 23 temporal bone specimens from 19 deceased donors with LO associated with the RWM) and the control group (comprising 27 age-matched specimens from 20 deceased donors without any otologic diseases). In the LO group, the 19 donors included 11 men and 8 women; their mean age was 56.42 ± 22.62 years (range, 3-94 years). In the control group, the 20 donors included 12 men and 8 women; their mean age was 53.95 ± 16.66 years (range, 16-77 years).

In the LO group, we found bilateral evidence of ossification in specimens from 4 (17%) donors and unilateral evidence in 15 (83%). We saw ossification in specimens from 5 donors with chronic otitis media, 4 with otogenic meningitis, 2 with leukemia, 1 with trauma, 1 with surgical removal of acoustic neuroma, 1 with polyarteritis nodosa, 1 with Fabry disease, 1 with cancer metastases to the temporal bone, 1 with systemic lupus erythematosus, and 1 with an autoimmune inner ear disease (Figure 1). We had no medical record for 1 donor.

In both groups, we documented the location of LO in the cochlea, the intensity of endolymphatic hydrops, the number of spiral ganglion cells and cochlear hair cells, and the areas of the stria vascularis and spiral ligament. Then, we compared our findings between the 2 groups. In addition, we created a new pathologic grading system for temporal bone specimens from deceased donors with LO associated with the RWM.

**Endolymphatic Hydrops**

We subdivided our temporal bone specimens according to the intensity of hydrops, per the classification of Cureoglu et al.: slight hydrops—bulging of Reissner’s membrane without contact with the bony wall of the scala vestibuli; moderate hydrops—displacement of Reissner’s membrane with contact with the wall of the scala vestibuli but with an angle with the osseous spiral lamina <90 degrees; and profound hydrops—displacement of Reissner’s membrane with bony contact, with an angle with the osseous spiral lamina ≥90 degrees.

**Spiral Ganglion Cells**

We divided Rosenthal’s canal into 4 segments, as described previously: segment I (from the base to 6 mm), segment II (6-15 mm), segment III (15-22 mm), and segment IV (22 mm to the apex). In each section, we counted all nuclei. To determine the number of ganglion cells in each segment and in the cochlea as a whole, we multiplied their summed counts by 10 (to account for the unmounted sections) and by a factor of 0.9 (to account for cells located at the interface between sections).

**Cochlear Hair Cells**

In each section, we counted the number of present and missing cochlear hair cells. In all turns of the cochlea, we calculated the percentage of hair cell loss by dividing the number of missing hair cells by the total number of hair cells possible in that turn.

**Stria Vascularis**

In all turns of the cochlea at the midmodiolar level, as well as on the adjacent 2 sections, we obtained morphometric measurements of the area of the stria vascularis. We acquired each image with a digital camera (at a magnification ×200). Using a computer, we quantified the areas of the cut surfaces of the stria vascularis. For the measurements, we used commercially available image analysis software (SPOT Advanced; SPOT Imaging Solutions, Sterling Heights, Michigan). Excluded from the area of the stria vascularis were any secondary changes, such as cystic-like structural areas or concretions.

**Spiral Ligament**

In all turns of the cochlea at the midmodiolar level, as well as on the adjacent 2 sections, we obtained morphometric measurements of the area of the spiral ligament. We acquired each calibrated image with a charge-coupled device camera (at a magnification ×40) connected to a personal computer. Using that computer, we quantified the areas of the cut surfaces of the spiral ligament. For the measurements, we used commercially available image analysis software (SPOT Advanced).
Our Grading System

We created a new pathologic grading system for temporal bone specimens from deceased donors with LO associated with the RWM (Figure 2). The 4 grades that we devised, according to the location and extent of ossification, are as follows:

- **Grade I**: Ossification involves a part of the RWM with or without scattered calcification in the scala tympani.
- **Grade II**: Ossification involves the whole RWM with or without less than half of the scala tympani.
- **Grade III**: Ossification involves the whole RWM, and half of the scala tympani or more than half of the scala tympani.
- **Grade IV**: Ossification involves the whole RWM, more than half of the scala tympani, and at least 1 other scala (the scala vestibuli and/or the scala media).

Statistical Analysis

To analyze any differences between the LO group and the control group, we used the Mann-Whitney U test. Significance was defined as $P < .05$.

Results

Location

For the 23 temporal bone specimens in the LO group, the location of LO within the inner ear is listed in the Table 1. We most frequently observed LO in the scala tympani of the basal turn of the cochlea (in 74% of the specimens).

Endolymphatic Hydrops

We found a significant difference between the 2 groups in the intensity of endolymphatic hydrops ($P < .001$). We could not evaluate 7 specimens in the LO group (Figure 1). Of the remaining 16 specimens in the LO group, 9 (56%) showed hydrops (3 profound, 2 moderate, and 4 slight). In contrast, of the 27 specimens in the control group, only 1 (3%) showed hydrops (slight).

Table 1. Location of Labyrinthitis Ossificans in the Inner Ear.

<table>
<thead>
<tr>
<th>Location</th>
<th>Temporal Bones, n</th>
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<tbody>
<tr>
<td>Semicircular canal</td>
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<tr>
<td>Superior</td>
<td>4</td>
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<tr>
<td>Lateral</td>
<td>12</td>
</tr>
<tr>
<td>Posterior</td>
<td>13</td>
</tr>
<tr>
<td>Utricle</td>
<td>8</td>
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<tr>
<td>Saccule</td>
<td>8</td>
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<tr>
<td>Basal turn</td>
<td></td>
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<tr>
<td>Scala vestibuli</td>
<td>10</td>
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<tr>
<td>Scala media</td>
<td>7</td>
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<tr>
<td>Scala tympani</td>
<td>17</td>
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<tr>
<td>Middle turn</td>
<td></td>
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<tr>
<td>Scala vestibuli</td>
<td>6</td>
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<tr>
<td>Scala media</td>
<td>4</td>
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<tr>
<td>Scala tympani</td>
<td>6</td>
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<tr>
<td>Apical turn</td>
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<tr>
<td>Scala vestibuli</td>
<td>4</td>
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<td>Scala media</td>
<td>4</td>
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<tr>
<td>Scala tympani</td>
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Spiral Ganglion Cells

We found a significant difference in the total number of spiral ganglion cells between the LO group (14,296) and the control group (20,022; $P = .001$). We also found a significant difference in the number of spiral ganglion cells in segment I between the LO group (1985) and the control group (2729; $P = .006$), in segment II between the LO group (5194) and the control group (7717; $P = .001$), in segment III between the LO group (3252) and the control group (4427; $P = .002$), and in segment IV between the LO group (3864) and the control group (5148; $P = .007$; Figure 3).

Cochlear Hair Cells

We found a significant difference between the 2 groups in the loss of outer hair cells in the lower ($P < .001$) and upper ($P = .001$) cochlear basilar turn, in the lower ($P = .007$)
and upper \((P = .001)\) cochlear middle turn, and in the apical \((P = .032)\) cochlear turn (Figure 3). We also found a significant difference between the 2 groups in the loss of inner hair cells in the lower \((P = .005)\) and upper \((P = .004)\) cochlear basal turn, in the lower \((P = .033)\) and upper \((P = .001)\) cochlear middle turn, and in the apical \((P = .009)\) cochlear turn (Figure 3).

**Stria Vascularis**

We found a highly significant difference \((P < .001)\) between the 2 groups in the presence of atrophy of the stria vascularis in all turns of the cochlea (lower basal, upper basal, lower middle, upper middle, and apical; Figure 4).

**Spiral Ligament**

We found a significant difference between the 2 groups in the presence of atrophy of the spiral ligament in the lower \((P = .002)\) and upper \((P < .001)\) cochlear basal turn, as well as in the lower \((P = .028)\) and upper \((P = .016)\) cochlear middle turn but not in the apical cochlear turn \((P > .05)\;\text{Figure 5.}\)

**Our Grading System**

Of the 23 temporal bone specimens in the LO group, 10 (43%) had grade I ossification; 5 (22%), grade II ossification; 1 (4%), grade III ossification; and 7 (31%), grade IV ossification.

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**Figure 3.** Bar graphs of sensorineural elements: (A) mean number of the spiral ganglion cells by segment, (B) mean loss of OHCs, and (C) mean loss of IHCs. Error bars indicate standard deviations. IHCs, inner hair cells; LO, labyrinthitis ossificans; OHCs, outer hair cells. *\(P < .05\).*

**Figure 4.** Bar graph of mean area of the stria vascularis \((\mu m^2)\). Error bars indicate standard deviations. LO, labyrinthitis ossificans. *\(P < .05\).*

**Figure 5.** Bar graph of mean area of the spiral ligament \((\mu m^2)\). Error bars indicate standard deviations. LO, labyrinthitis ossificans. *\(P < .05\).*
Discussion

Bacteria and toxins may reach the inner ear typically through either the RWM ( tympanic origin) or the internal auditory canal and cochlear aqueduct (meningeal origin).1,8 In our study of 23 temporal bone specimens from deceased donors with LO associated with the RWM, 6 (26%) specimens showed chronic otitis media and 4 (17%), otogenic meningitis. After involvement of bacteria in the acute stage, fibrous and ossification stages can follow.1

Inflammatory products can lead to imbalance in the osmotic pressure between the perilymph and the endolymph due to alteration in electrolytes and proteins.1 Those changes in ion fluid and protein balance may explain the development of endolymphatic hydrops in patients with LO.19 In our study, we found that the intensity of endolymphatic hydrops was significantly increased in the LO group as compared with the control group—a finding consistent with previously reported data.1,10

In patients with profound deafness, various diseases, including bacterial and viral labyrinthitis, lead to the loss of spiral ganglion cells.11 Although the good preservation of several cochlear structures of the labyrinth may affect the success of the cochlear implant surgery, most important seems to be a sufficient number of spiral ganglion cells.7,12,13 A negative correlation has been observed between the duration of deafness and the number of spiral ganglion cells.14 In addition, Suga and Lindsay demonstrated a wide variation of histopathologic changes in temporal bone specimens from patients with LO, including damage to sensorineural structures.2 Similarly, in an animal study, Tinling et al showed that cochlear damage caused by LO was mostly in the basal turn and was reduced toward the apical turn.15 They observed that cochlear elements degenerated even in the absence of ossification in the adjacent scala tympani or scala vestibuli. In our study, we found a significant loss in the number of outer hair cells, inner hair cells, and spiral ganglion cells in all segments of the temporal bone specimens with LO. Our findings suggest that cochlear hair cells might be very sensitive to inflammatory changes occurring within the other cochlear turns.15

Several previous studies have revealed degeneration to the areas of the stria vascularis and spiral ligament in gerbil15 and human2,16 temporal bones with pathologic new bone formation. In our study, in the LO group, we observed a significant decrease in the areas of the stria vascularis in all turns of the cochlea in LO, as well as a significant decrease in the areas of the spiral ligament in the basal and middle cochlear turn. In such donors, ischemia due to vascular occlusion from inflammation might be the cause of cochlear lateral wall changes.1

LO is known to be the most frequent (13%) abnormality in cochlear implant candidates.17 In our study, we found that—regardless of the cause of ossification—the most common location for LO was in the scala tympani of the basal turn of the cochlea (74%), a finding consistent with previous reports.10

A goal during cochlear implant surgery is to insert an electrode array into the scala tympani through the RWM. Extensive ossification and/or fibrosis into the cochlea around the RWM can render such surgery difficult or impossible. Most inflammatory products use the RWM and/or the cochlear aqueduct to enter the inner ear, so pathologic bone growth most commonly occurs in the scala tympani near the RWM.1,5 According to our new pathologic grading system, based on the location and extent of ossification associated with the RWM, 8 (35%) temporal bone specimens were either grade III or grade IV—suggesting that severe ossification around the RWM is common. Therefore, once a patient is diagnosed with sensorineural hearing loss associated with diseases that can cause LO, cochlear implant surgery should be performed as soon as possible. Unfortunately, given the limited availability of clinical records for the deceased donors in our study’s LO group, we were not able to determine the duration of ossification in our donors.

Conclusion

Our quantitative histopathologic analysis of temporal bone specimens from deceased donors with LO associated with the RWM (as compared with the control group) revealed a significantly increased intensity of endolymphatic hydrops; a significant degeneration in the spiral ganglion cells in all segments; a significantly higher loss of outer and inner hair cells in all turns of the cochlea; a significantly increased degree of atrophy of the area of the stria vascularis in all turns of the cochlea; and a significantly increased degree of atrophy of the area of the spiral ligament in the basal and middle cochlear turn. Timely cochlear implant surgery is very important: it should be performed as soon as possible after the diagnosis of sensorineural hearing loss associated with diseases that can cause LO.

Author Contributions

Serdar Kaya, conception and design of study, acquisition, analysis, and interpretation of data, drafting and revising manuscript, final approval of manuscript, and agreement to be accountable for all aspects of work; Michael M. Paparella, conception of study, analysis and interpretation of data, critical revision of manuscript, final approval of manuscript, and agreement to be accountable for all aspects of work; Sebahattin Cureoglu, conception and design of study, acquisition, analysis and interpretation of data, drafting and revising manuscript, final approval of manuscript, and agreement to be accountable for all aspects of work.

Disclosures

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Sponsorships: None.

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References


