

Effects of bone morphogenetic protein 4 on differentiation of embryonic stem cells into myosin VIIa-positive cells

TORU HIGASHI¹, TAKAYUKI NAKAGAWA¹, TOMOKO KITA¹, TAE-SOO KIM¹,
TATSUNORI SAKAMOTO², & JUICHI ITO¹

¹Department of Otolaryngology-Head and Neck Surgery, Kyoto University Graduate School of Medicine, Kyoto and

²Organogenesis and Neurogenesis Group, Center for Developmental Biology, Riken, Kobe, Japan

Abstract

Conclusion: Our results indicate that myosin VIIa-positive cells are generated from embryonic stem cells (ESCs) co-cultured with PA6 cells; however, bone morphogenetic protein 4 (BMP4) may not be a key molecule for induction of myosin VIIa-positive cells from the ESCs. **Background:** ESCs have been considered as a basis for cell therapy in a range of organs, because of their potential for self-renewal and pluripotency. Co-culture with PA6 stromal cells can induce differentiation of ESCs into various types of ectodermal cells including sensory progenitors. BMP4 plays an essential role in the development of sensory hair cells in the inner ear. **Materials and methods:** We examined effects of BMP4 on differentiation of ESCs into the hair cell immunophenotype. BMP4 was supplemented at different time points to ESCs co-cultured on PA6 stromal cells. The ESCs were then collected and examined for the expression of myosin VIIa, a hair cell marker, and β III-tubulin, a neural marker. The expression of myosin VIIa and β III-tubulin was identified. **Results:** Quantitative assessments revealed that exogenous BMP4 has significant effects on the expression of β III-tubulin, but not of myosin VIIa.

Introduction

Bone morphogenesis protein 4 (BMP4) plays essential roles in the development of the inner ear including sensory epithelium [1–3]. Recently, it has been reported that BMP regulates differentiation and cell proliferation in developing auditory epithelia of chicks [4]. Hair cell regeneration has been a central issue in the field of inner ear research for decades. However, hair cell regeneration in mammalian cochleae is a problem that is still being discussed. Recent studies using gene transfer by adenovirus vectors have demonstrated the potential of supporting cells for transdifferentiation into hair cells [5,6]. However, in severely damaged cochleae, there are no remaining supporting cells that are capable of transdifferentiation into hair cells. In such circumstances, cell transplantation may be a possible strategy for hair cell regeneration.

Embryonic stem cells (ESCs) have been considered as a basis for cell therapy in a range of organs, because of their capability for self-renewal and

pluripotency. Recently, it has been reported that ESCs can differentiate into inner ear cells including sensory hair cells [7]. We have also examined the potential of ESCs as a source of transplants for regeneration of spiral ganglion neurons [8,9]. ESC-derived cells following induction of differentiation into ectodermal cells are desirable, because most inner ear cells are originated from the ectoderm. Several methods for ectodermal induction of ESCs have been established. We have used the stromal cell-inducing activity (SDIA), in which ESCs are co-cultured with PA6 cells [10], stromal cells derived from mouse skull bones, for this purpose [8,9]. SDIA treatment can induce various types of neuronal cells with a combination of supplements of BMP4 or sonic hedgehog [11]. However, the potential of SDIA treatment for induction of inner ear sensory cells has not been examined. The aim of this study was set to determine the effects of BMP4 on differentiation of SDIA-treated ESCs into inner ear sensory cells. The profile of differentiation of SDIA-

treated ESCs by exposure to BMP4 at various time points was analyzed by immunocytochemistry.

Materials and methods

Materials

Mouse G4-2 ESCs (donated by Hitoshi Niwa of Riken CDB, Kobe, Japan) derived from the E14tg2a ESC line [12], and carrying the enhanced green fluorescence protein (EGFP) gene driven by the CAG promoter, were used in this study. The PA6 cells (RCB1127), a stromal cell line derived from newborn mouse calvaria were provided from Riken Cell Bank, Kobe, Japan. Glasgow's Modified Eagle's Medium (GMEM), knockout serum replacement (KSR), and nonessential amino acids (NEAA) were purchased from Invitrogen (Carlsbad, CA, USA), pyruvate was from Sigma (St Louis, MO, USA) and 2-mercaptoethanol (2-ME) was from Wako, Osaka, Japan. Recombinant human BMP4 was from R&D Systems (Cleveland, OH, USA).

Induction of differentiation of ESCs

We used the SDIA for neural induction of ESCs [10,11]. ESCs were cultured to form differentiated colonies on a feeder layer of PA6 stromal cells derived from newborn mouse calvaria in GMEM supplemented with 5% KSR, 1 mM pyruvate, 0.1 mM NEAA, and 0.2 mM 2-ME at 37°C in a 5% CO₂ atmosphere. We set six conditions for the culture of ESCs during SDIA treatment according to the duration of the exposure to BMP4 at a concentration of 0.5 nM; during day 1–6, 2–6, 3–6, 4–6, 5–6 or no exposure. Colonies that formed on the PA6 monolayer after 6 days of culture were isolated by collagenase B (Roche Diagnostics, Tokyo, Japan). Cell suspensions of SDIA-treated ESCs were adjusted to a concentration of 10³ cells/μl in GMEM, and a 10 μl portion of the cell suspension was then replaced onto a sterile membrane (Falcon™ Cell Culture Insert; 3.0 μm pore size, 24-well format; Becton Dickinson Labware, Franklin Lakes, NJ, USA) in a 24-well culture plate (Asahi Techno Glass Corp., Tokyo, Japan). SDIA-treated ESCs were incubated in the culture medium containing no BMP4 for an additional 7 days. Each experimental condition consisted of five wells.

Immunocytochemistry

Following incubation, the cultured specimens were washed with phosphate-buffered saline (PBS; Nacalai Tesque Inc., Kyoto, Japan) and fixed with 4% paraformaldehyde in PBS for 15 min. The fates of cultured ESCs were determined by immunostaining

for myosin VIIa, a marker for hair cells, Pax-2, a marker for sensory progenitors in the inner ear [13], and βIII-tubulin in whole mounts. ESC-derived cells obtained from four wells in each culture condition were double-stained with rabbit anti-myosin VI (×700; purchased from Tama Hasson, University of California, San Diego, CA, USA), and mouse anti-βIII-tubulin (×200; Covance, Berkeley, CA, USA). Secondary antibodies used were Alexa-Fluor 546-conjugated anti-rabbit and Alexa-Fluor 633-conjugated anti-mouse goat antibodies (×200; Molecular Probes, Eugene, OR, USA). Immunocytochemistry for rabbit anti-Pax-2 (×200; Covance) was performed in one well for each condition. The specimens were viewed with a Leica TCS SP2 confocal laser scanning microscope (Leica Microsystems Inc., Wetzlar, Germany). In each well, we counted the numbers of ESC-derived colonies labeled by EGFP and those of colonies containing myosin VIIa- or βIII-tubulin-positive cells. The ratio for each marker-expressing colony was then calculated. The differences in the ratio for each marker among culture conditions were statistically analyzed by ANOVA with Fisher's protected least significant difference (PLSD). A *p* value <0.05 was considered significant.

Results

Both βIII-tubulin- and myosin VIIa-positive colonies were found in all the culture conditions. In these colonies, the majority of myosin VIIa-positive cells were located in the central portion of the colonies surrounded by βIII-tubulin-positive cells (Figure 1A–C). A few myosin VIIa-positive cells were found in the peripheral lesion of the colony. Immunostaining for βIII-tubulin demonstrated massive elongation of neurites from ESC-derived cells (Figure 1D–F). On the other hand, we found no cells that exhibited the expression of both myosin VIIa and βIII-tubulin. We found one or two ESC-derived colonies containing Pax-2-positive cells in cultures that were exposed to BMP4 during day 2–6, day 5–6 or no exposure (Figure 2).

Quantitative assessment for the ratio of βIII-tubulin-positive colonies demonstrated significant effects of BMP4 application on differentiation of ESCs into neurons (*p*=0.0002; Figure 3A). The differences in the ratio of βIII-tubulin-positive colonies between day 1–6 or 2–6 and day 4–6, 5–6 or no exposure, and between day 3–6 and day 5–6 or no exposure were significant at Fisher's PLSD. Early exposure to BMP4 reduced the ratio for βIII-tubulin-positive colonies. On the other hand, BMP4 application had no significant effect on the ratio for myosin VIIa-positive colonies (Figure 3B).

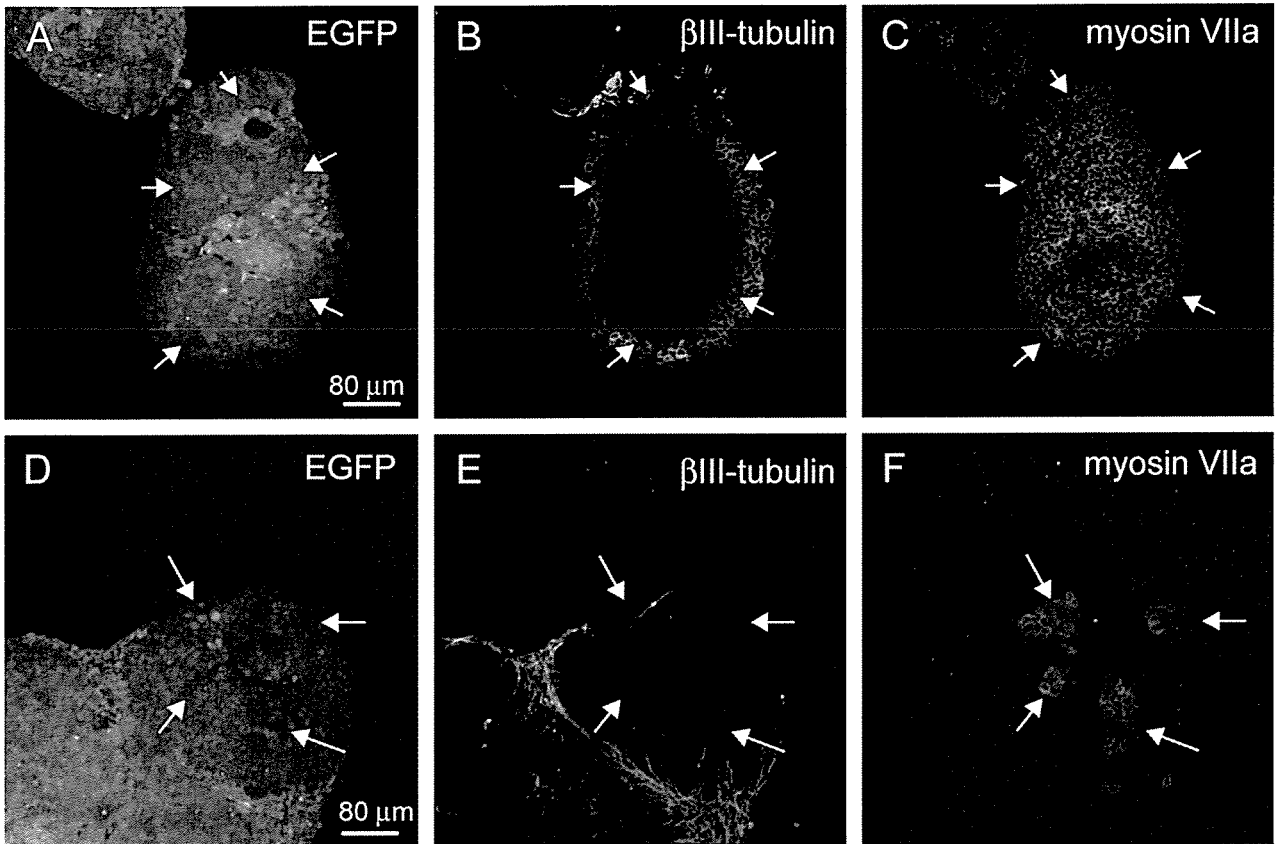


Figure 1. Expression of β III-tubulin and myosin VIIa in SDIA-treated embryonic stem cells. (A–C) Embryonic stem cell colony exposed to BMP4 during day 3–6. (D–F) Embryonic stem cell colony cultured without exposure to BMP4. Arrows indicate the location of myosin VIIa-positive cells.

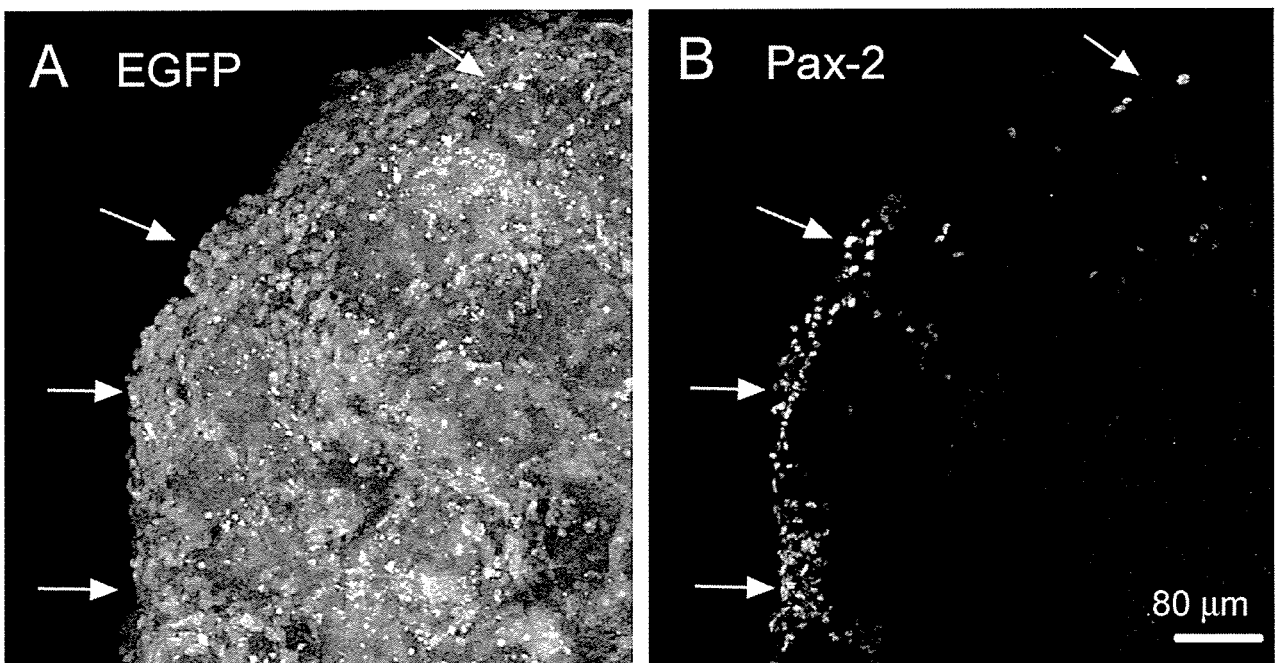


Figure 2. Expression of Pax-2 in SDIA-treated embryonic stem cells. Some embryonic stem cells exposed to BMP4 during day 2–6 exhibit the expression of Pax-2 (arrows).

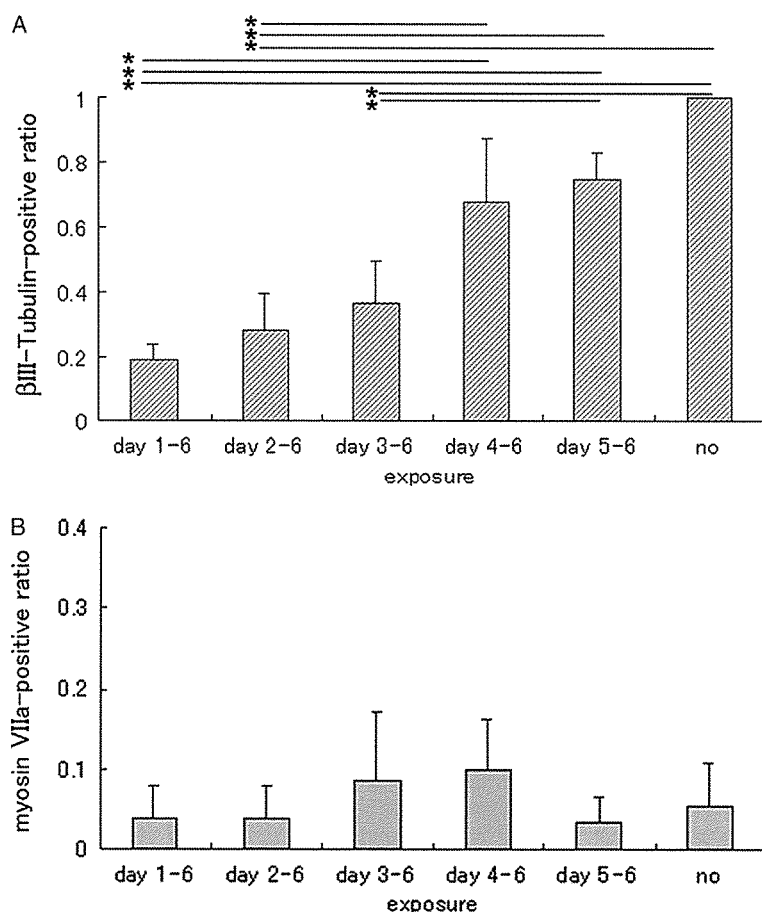


Figure 3. Quantitative assessment of the expression of β III-tubulin and myosin VIIa in colonies of embryonic stem cells. X-axis shows the duration of exposure to BMP4. Exposure to BMP4 has significant effects on the expression of β III-tubulin (A), and asterisks indicate significant differences in pairwise comparison. No significant differences were found in the ratio for myosin VIIa expression (B).

In culture conditions exposed to BMP4 from day 3 or 4 to day 6, the ratios for myosin VIIa expression were slightly higher than other culture conditions; however, no significant differences were found among experimental groups.

Discussion

Inner ear cell progenitors are derived from the ectoderm similar to neural or epidermal progenitors. The inner ear is derived from a thickened patch of ectodermal cells, which develops lateral to the developing hindbrain. The SDIA can generate various types of ectodermal cells from ESCs [11]. The present findings demonstrate that myosin VIIa- or Pax-2-positive cells can be derived from SDIA-treated ESCs, which indicates that hair cells may be generated from SDIA-treated ESCs.

Early exposure of SDIA-treated ESCs to BMP4 induces epidermal differentiation of ESCs, while no exposure to BMP4 during the SDIA treatment results in highly neural differentiation of ESCs [11]. In addition, sensory progenitors are generated

from SDIA-treated ESCs by the late exposure to BMP4 [11]. We therefore expected that late exposure of SDIA-treated ESCs to BMP4 might have the activity for differentiation of SDIA-treated ESCs into inner ear cells including hair cells. The present findings demonstrate that BMP4 has a significant effect on suppression of neural differentiation of SDIA-treated ESCs, which is identical to previous findings [11], indicating that the exposure to BMP4 certainly works in our culture systems. However, our results indicate no significant effects of BMP4 exposure on the expression of myosin VIIa in SDIA-treated ESCs. In addition, Pax-2-positive cells were generated with or without exposure to BMP4. These findings indicate that BMP4 exposure has no effects on differentiation of SDIA-treated ESCs into inner ear hair cells.

Recent studies on the development of the inner ear have indicated that BMP4 plays a crucial role in maturation of inner ear sensory epithelia [1–3]. In the chick otocyst culture system, BMP4 induces differentiation of hair cells from their progenitors, and promotes down-regulation of Pax-2 protein in

sensory epithelial progenitors, leading to reduced progenitor cell population [4]. Based on these findings, the early exposure to BMP4 might induce reduction of hair cell progenitors derived from ESCs, and the late exposure to BMP4 might induce differentiation of hair cells from the progenitors derived from ESCs. However, our findings demonstrated no down-regulation of Pax-2 in ESC-derived cells by the early exposure to BMP4 and no up-regulation of myosin VIIa by the late exposure. Therefore, the molecules that determine the fate of ESCs to differentiate into inner ears in the earlier stage of development may be required for induction of differentiation of ESCs into inner ear cells.

In conclusion, myosin VIIa-positive cell populations were obtained from SDIA-treated ESCs; however, no significant effects of BMP4 exposure on the expression of myosin VIIa in ESC-derived cells were identified. Further studies are required for identification of key molecules for induction of differentiation of SDIA-treated ESCs into hair cells.

Acknowledgements

We thank H. Niwa for providing ESCs, T. Hasson for providing the antibody for myosin VIIa, S. Sasai for providing PA6 cells, and R. Sadato and Y. Nishikawa for technical assistance. This study was supported by a Grant-in-Aid for the Regenerative Medicine Realization project from the Ministry of Education, Science, Sports, Culture and Technology of Japan.

References

- [1] Oh SH, Johnson R, Wu DK. Differential expression of bone morphogenetic proteins in the developing vestibular and auditory sensory organs. *J Neurosci* 1996;16:6463–75.
- [2] Takemura T, Sakagami M, Takebayashi K, Umemoto M, Nakase T, Takaoka K, et al. Localization of bone morphogenetic protein-4 messenger RNA in developing mouse cochlea. *Hear Res* 1996;95:26–32.
- [3] Cole LK, Le Roux I, Nunes F, Laufer E, Lewis J, Wu DK. Sensory organ generation in the chicken inner ear: contributions of bone morphogenetic protein 4, serrate1, and lunatic fringe. *J Comp Neurol* 2000;424:509–20.
- [4] Li H, Corrales CE, Wang Z, Zhao Y, Wang Y, Liu H, et al. BMP4 signaling is involved in the generation of inner ear sensory epithelia. *BMC Dev Biol* 2005;5:16.
- [5] Kawamoto K, Ishimoto S, Minoda R, Brough DE, Raphael Y. Math1 gene transfer generates new cochlear hair cells in mature guinea pigs in vivo. *J Neurosci* 2003;23:4395–400.
- [6] Izumikawa M, Minoda R, Kawamoto K, Abrashkin KA, Swiderski DL, Dolan DF, et al. Auditory hair cell replacement and hearing improvement by Atoh1 gene therapy in deaf mammals. *Nat Med* 2005;11:271–6.
- [7] Li H, Roblin G, Liu H, Heller S. Generation of hair cells by stepwise differentiation of embryonic stem cells. *Proc Natl Acad Sci U S A* 2003;100:13495–500.
- [8] Matsumoto M, Nakagawa T, Higashi T, Kim TS, Kojima K, Kita T, et al. Innervation of stem cell-derived neurons into auditory epithelia of mice. *Neuroreport* 2005;16:787–90.
- [9] Okano T, Nakagawa T, Endo T, Kim TS, Kita T, Tamura T, et al. Engraftment of embryonic stem cell-derived neurons into the cochlear modiolus. *Neuroreport* 2005;16:1919–22.
- [10] Kawasaki H, Mizuseki K, Nishikawa S, Kaneko S, Kuwana Y, Nakanishi S, et al. Induction of midbrain dopaminergic neurons from ESCs by stromal cell-derived inducing activity. *Neuron* 2000;28:31–40.
- [11] Mizuseki K, Sakamoto T, Watanabe K, Muguruma K, Ikeya M, Nishiyama A, et al. Generation of neural crest-derived peripheral neurons and floor plate cells from mouse and primate embryonic stem cells. *Proc Natl Acad Sci U S A* 2003;100:5828–33.
- [12] Hooper M, Hardy K, Handyside A, Hunter S, Monk M. HPRT-deficient (Lesch-Nyhan) mouse embryos derived from germline colonization by cultured cells. *Nature* 1987;326:292–5.
- [13] Lawoko-Kerali G, Rivolta MN, Holley M. Expression of the transcription factors GATA3 and Pax2 during development of the mammalian inner ear. *J Comp Neurol* 2002;442:378–91.

Bilateral congenital ossicular chain disruption mimicking otosclerosis

KIYOHITO FUJINO, SHIN-ICHI KANEMARU, HARUKAZU HIRAUMI & JUICHI ITO

Department of Otolaryngology-Head and Neck Surgery, Graduate School of Medicine, Kyoto University, Kyoto, Japan

Abstract

A rare case of bilateral congenital ossicular chain disruption whose history and findings mimicked those of otosclerosis is reported. A 54-year-old male consulted us for slowly progressing hearing loss. Audiogram showed bilateral intermediate mixed hearing loss. The stapedial reflex was negative and the tympanogram was normal A-type. Based on the diagnosis of otosclerosis, stapes surgery was planned for the left side. Surgical findings revealed normal mobility of the stapes and a small disruption at the incudostapedial joint with connection of intact mucosa. The disruption was repaired with auto-bone columella. The patient's hearing improved after surgery. The surgery for the other side was performed successively, and similar ossicular disruption was observed. The mechanisms of findings that misled the preoperative diagnosis are discussed.

Introduction

Disruption of the ossicular chain is caused by trauma, cholesteatoma, or congenital malformation. Diagnosis of this disorder is usually not difficult from the history and the otological test findings. We experienced a rare case of bilateral congenital ossicular disruption whose history and findings mimicked those of otosclerosis. We planned a stapes surgery for the left side, but we found a small defect of the incus near the incudostapedial joint during surgery. The disruption was repaired, and the patient's hearing improved. Surgery for the other side revealed the same type of disruption.

Case report

A 54-year-old male consulted us for slowly progressing bilateral hearing loss over a period of several years. He did not have any history of head injury or otitis media. His ear drums were normal. Pure tone audiogram (PTA) showed bilateral moderate mixed hearing loss (Figure 1). The stapedial reflex was negative on both sides. Tympanogram showed normal A-type (not Ad- or As-type). CT scan showed no evidence of ossicular malformation or disruption. Based on the history

and findings, we diagnosed this case as bilateral otosclerosis. We first planned a stapes surgery for the left side. Surgery was performed by a trans-meatal approach. Unexpectedly we found that the mobility of the stapes was normal. The mobility of the malleus and incus was also normal. However, there was a small defect of the incus at the lenticular process. This part was connected only with the intact mucosa that covered the ossicles (Figure 2A). When we vibrated the incus with a Rosen needle, the vibrations were transmitted to the stapes at a very low rate. A small protuberance on the long crus of the incus just above the discontinued part was noted as combined malformation. The surgical procedure was changed to the repair of ossicular disruption. The mucosa that covered the defect was cut, and a small particle of the external canal bone was interposed into the discontinued part, and was fixed with fibrin glue (Figure 2B). After surgery, the PTA threshold improved from 63.3 to 45.0 dB (average of 0.5, 1, and 2 kHz; Figure 3). Three months after the surgery for the left side, surgery for the right side was performed. Surgical findings were almost the same as those in the left side. The ossicular disruption was repaired in the same way.

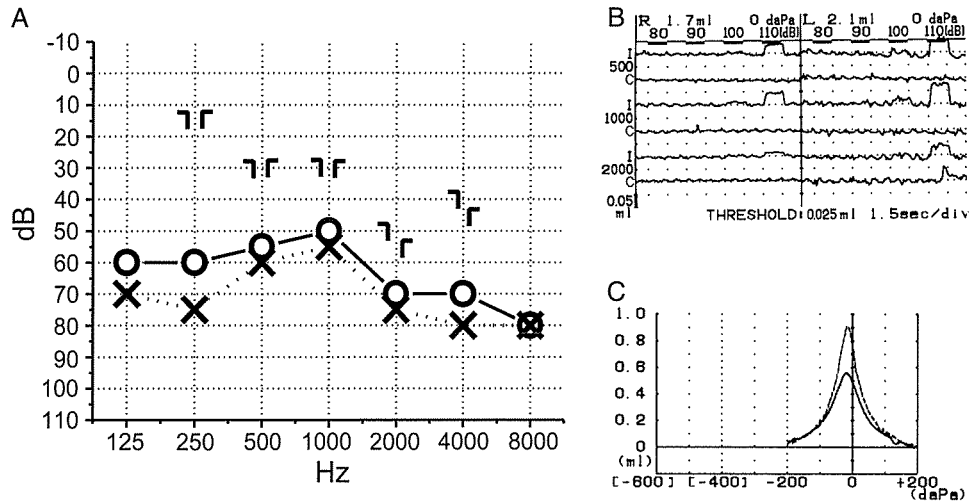


Figure 1. Preoperative otological test findings. (A) Pure tone audiogram (PTA) showed moderate mixed hearing loss on both sides. Average (0.5, 1, and 2 kHz) air conduction thresholds/air–bone gaps were: right 58.3/21.7 dB, left 63.3/25 dB. (B) The stapedial reflex was negative on both sides. Opposite (upward) responses were seen in some parts. (C) The tympanogram showed normal A-type on both sides. The solid line indicates the right side, and the dashed line indicates the left side.

Discussion

Since the patient did not have any history of trauma or otitis media, we consider the cause of ossicular disruption to be congenital malformation. The patient complained of progressive hearing loss over a period of several recent years. The reason for such a subjective symptom is not clear. The patient may have had mild hearing loss due to ossicular discontinuity since his childhood. The recent hearing loss may have been due to aggravation of the sensorineural component by presbycusis. The ossicular

disruption usually demonstrates a large air–bone gap of PTA, negative stapedial reflex, and Ad-type tympanogram [1–3]. In our case findings were different except for the stapedial reflex, probably because the discontinued part was very short and was connected with mucosa. Preoperative CT scan could not detect the discontinuity because the defect was too small for the resolution.

There have been several reports of congenital disconnection of the incudostapedial joint [4–6]; however, we could not find a case in which the

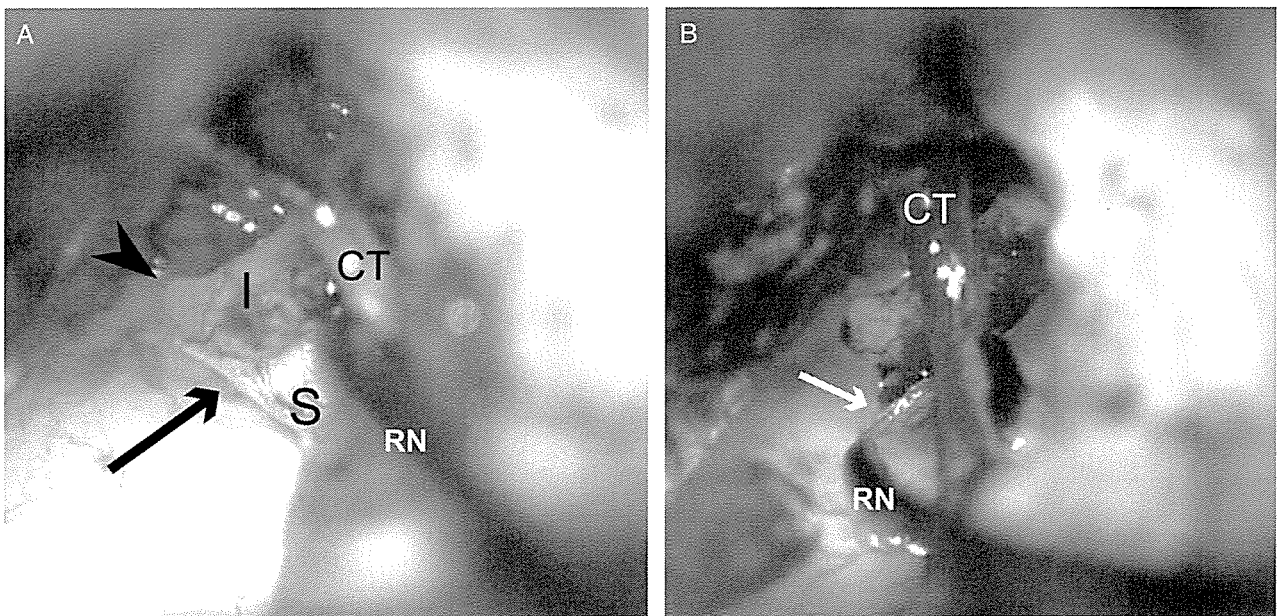


Figure 2. Findings during surgery of the left side. (A) A small pinched part near the incudostapedial joint was found (arrow). This part lacked bone and connected only with normal mucosa that covered the bone. A small protuberance on the incus just above the discontinued part was noted as combined malformation (arrowhead). (B) A particle of external canal bone (arrow) was inserted into the discontinued part with a Rosen needle. I, long crus of incus; S, stapes head; CT, chorda tympani; RN, Rosen needle.

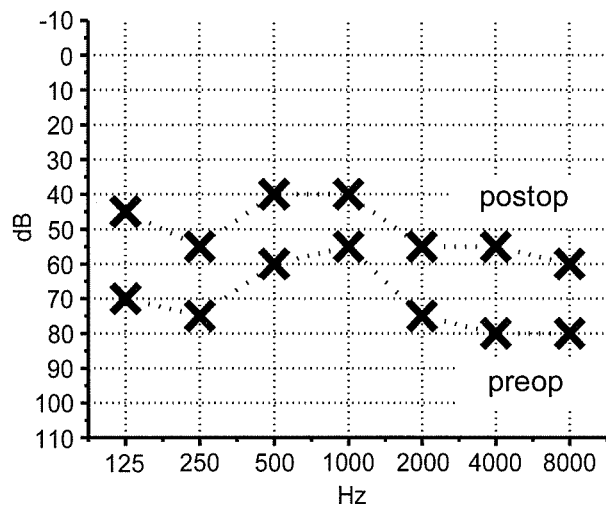


Figure 3. Pure tone audiogram of the left side examined before and 2 months after the surgery. Average air conduction threshold improved from 63.3 to 45.0 dB.

discontinued part was covered and connected with intact mucosa. Our case is rare in that the ossicular disruption was masked and appeared like otosclero-

sis because the length of the defect was short and it was connected with normal mucosa.

References

- [1] Liden G, Bjorkman G, Nyman H, Kunov H. Tympanometry and acoustic impedance. *Acta Otolaryngol (Stockh)* 1977;83: 140-5.
- [2] Liden G, Harford E, Hallen O. Tympanometry for the diagnosis of ossicular disruption. *Arch Otolaryngol* 1974;99: 23-9.
- [3] Daneshi A, Shafeghati Y, Karimi-Nejad MH, Khosravi A, Farhang F. Hereditary bilateral conductive hearing loss caused by total loss of ossicles: a report of familial expansile osteolysis. *Otol Neurotol* 2005;26:237-40.
- [4] Minatogawa T, Iritani H, Ishida K, Node MN. Allograft stapes surgery for conductive hearing loss in patients with ossicular chain anomalies. *Eur Arch Otorhinolaryngol* 1996; 253:283-6.
- [5] Bess FH, Miller GW, Glasscock ME 3rd, Bratt GW. Unexplained conductive hearing loss. *South Med J* 1980;73: 335-8.
- [6] Funasaka S. Congenital ossicular anomalies without malformations of the external ear. *Eur Arch Otorhinolaryngol* 1979; 224:231-40.

Stabilization technique for columella using trimmed autologous temporal fascia in type III and IV tympanoplasty – muffler method

SHIN-ICHI KANEMARU¹, JUICHI ITO¹, JUN TSUJI¹, KIYOHICO FUJINO¹, HARUKAZU HIRAUMI¹, & KOICHI OMORI²

¹Department of Otolaryngology-Head and Neck Surgery, Graduate School of Medicine, Kyoto University, Kyoto and

²Department of Otolaryngology, Fukushima Medical University School of Medicine, Fukushima, Japan

Abstract

Conclusion: High success rates of recovery of hearing level in type III and IV tympanoplasty could be achieved by this stabilization technique for columella using trimmed autologous temporal fascia. **Objective:** The aim of this study was to evaluate a new stabilization technique for columella using trimmed autologous temporal fascia in type III and IV tympanoplasty. **Patients and methods:** A total of 55 patients (21 male, 34 female, aged 4–85 years) with chronic otitis media ($n = 16$) and cholesteatoma ($n = 39$) underwent tympanoplasty using this new stabilization technique for columella. Thirty-one patients underwent type III tympanoplasty and 24 patients underwent type IV tympanoplasty. Forty-two patients underwent a staged operation and 13 patients underwent a single operation. The observation period was 3.5 years from 6 months after the last operation. **Results:** The overall success rates in type III and IV tympanoplasty were 87.1% (27/31) and 83.3% (20/24), respectively. Two of eight patients for whom the procedure was unsuccessful underwent reoperation and they acquired good hearing.

Keywords: Reconstruction of sound conducting system, dislocation of columella, chronic otitis media, cholesteatoma

Introduction

Tympanoplasty is a well-established procedure for the reconstruction of the sound conducting system in middle ear disease. However, hearing results are not always satisfactory, especially in type III and/or IV tympanoplasty [1,2]. One of the major causes of this failure is the dislocation of substitute for ossicles [3,4]. We contrived a new stabilization technique for columella using trimmed autologous temporal fascia in type III and IV tympanoplasty and achieved better hearing results. This simple stabilization technique is detailed and illustrated in this clinical report.

columella during a 3.5 year period in our university hospital and another three general hospitals. The patients ranged in age from 4 to 85 years; 25 patients were male and 42 patients were female. Thirty-seven patients underwent type III tympanoplasty and 30 patients underwent type IV tympanoplasty. Partial or total canal wall down tympanomastoidectomy was done in 35 of 41 patients with cholesteatoma and canal wall reconstruction was done in 28 of these patients. Forty-nine patients underwent a staged operation and 18 patients underwent a single-stage operation. The observation period was 3.5 years from 6 months after the last operation.

Patients and methods

Patients and diseases

Sixty-seven patients with chronic otitis media ($n = 26$) and cholesteatoma ($n = 41$) underwent tympanoplasty using this new stabilization technique for

Operation technique

Hydroxylapatite total or partial ossicular replacement prosthesis (Apaceram, Pentax, Asahi Optical, Tokyo, Japan) was applied for reconstruction of the ossicular chain. An autologous trimmed cartilage

Correspondence: Shin-ichi Kanemaru, MD, PhD, Department of Otolaryngology and HNS, Graduate School of Medicine, Kyoto University, 54 Kawahara-cho, Syogoinn, Sakyo-ku Kyoto 606-8507 Japan. Tel: +81 75 751 3350. Fax: +81 75 751 7225. E-mail: kanemaru@ent.kuhp.kyoto-u.ac.jp

that was harvested from the auricle was glued on the head of the prosthesis. The entire unit was placed between the stapes or its footplate and the tympanic membrane. A harvested autologous temporal fascia was dried and was trimmed into a string (1.5–2.0 × 20 mm) (Figure 1). This string was put around the neck of the prosthesis like a muffler (Figure 2). Both ends of the string were attached to the surrounding bone walls (facial canal in most cases) by fibrin glue (Bolheal, Kaketsuken, Kumamoto, Japan) (Figure 3).

Results

Air–bone conduction thresholds were calculated as a postoperative pure tone average of three speech frequencies (0.5, 1, and 2 kHz). Hearing success was defined as an average air–bone gap of ≤20 dB. Overall success rates in type III and IV tympanoplasty were 83.8% (31/37) and 76.7% (23/30), respectively (Table I). Three of 13 patients who did not have improved postoperative hearing realized that sudden hearing loss happened immediately after strongly blowing their noses, sneezing and/or nasal sniffing while they were in the hospital. The cause of these sudden hearing losses was discovered to be the dislocation of columella by high-resolution CT imaging of the middle ear [4]. Two of these three patients underwent reoperation with the same technique and could achieve good hearing.

Discussion

The aim of tympanoplasty is not only removal of lesions but also hearing improvement. At the present

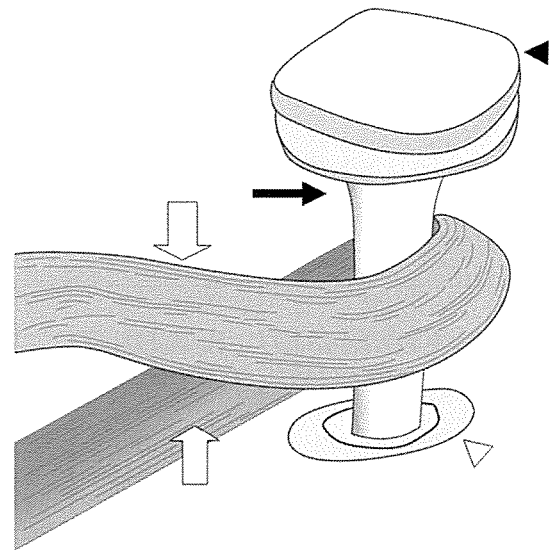


Figure 2. A schema of a temporal fascia string that was put around the neck of the prosthesis like a muffler. The black arrowhead indicates the autologous auricular cartilage, the black arrow indicates hydroxylapatite columella, the white arrowhead indicates the foot plate of the stapes, and the white arrows indicate the temporal fascia string (muffler).

time, however, satisfactory results of the latter have not been obtained in type III and IV tympanoplasty [1,2]. There are many causes of these failures; factors related to ossicular chains, immobility of the ossicles themselves and/or dislocation of columella are thought to be the main causes [3,4].

In type III and IV tympanoplasty, although it depends on the original diseases, an unstable

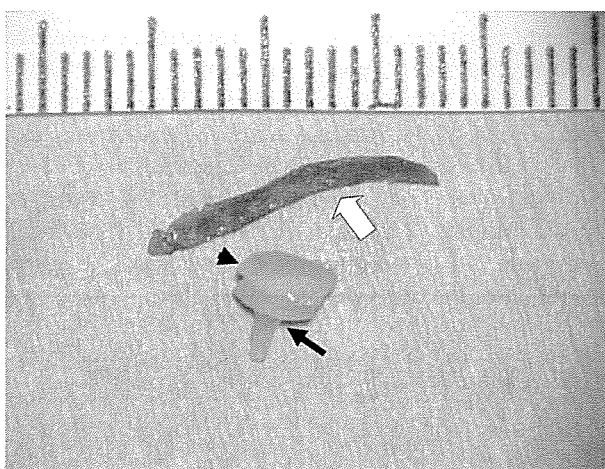


Figure 1. A hydroxylapatite partial ossicular replacement prosthesis with an autologous trimmed cartilage and an autologous temporal fascia that was dried and trimmed into a string. The black arrowhead indicates autologous auricular cartilage, the black arrow indicates hydroxylapatite columella, and the white arrow indicates the temporal fascia string (muffler).

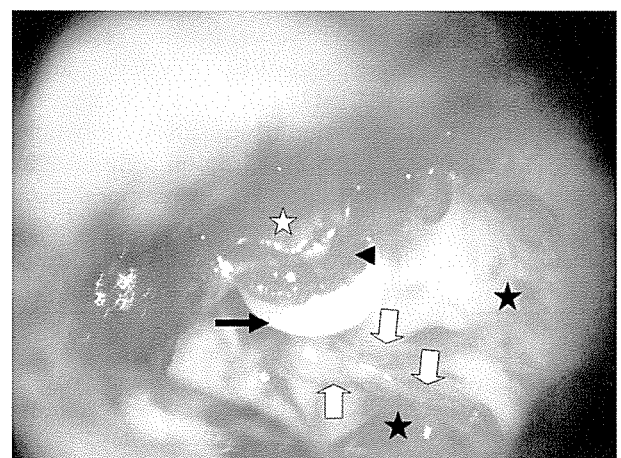


Figure 3. Reconstruction of the sound conductive system in tympanoplasty type IV. The black arrowhead indicates the autologous auricular cartilage, the black arrow indicates hydroxylapatite columella, the white arrows indicate the temporal fascia string (muffler), the black asterisks show the tympanic segment of the facial canal, and the white asterisk indicates the tympanic membrane.

Table I. Success rate of hearing and cause of failure in cases treated by the muffler method of tympanoplasty.

| | | | |
|-----------------------|---------------|--------------------------|---------------|
| Overall success rate* | 80.6% (54/67) | Tympanoplasty type III | 83.8% (31/37) |
| | | Tympanoplasty type IV | 76.7% (23/30) |
| Overall failure rate | 19.4% (13/67) | Dislocation of columella | 23.1% (3/13) |
| | | Unknown | 76.9% (10/13) |

*Hearing success is defined as a postoperative pure tone average (0.5 KHz, 1.0 KHz, and 2.0 KHz) air-bone gap of ≤ 20 dB.

columella has to be placed on the superstructure or the footplate of the stapes. There is no sustainable structure around the columella. To date, various devices and materials have been conceived and used to try and prevent dislocation of columella [3,5-7]. It is desirable that materials for stabilization of columella have the following characteristics: (1) no foreign body reaction, (2) small volume, and (3) no prevention of sound conduction. The autologous temporal fascia that we utilized in this study is an ideal material from the viewpoint of affinity and its volume is small enough to acquire the tympanic air space. Moreover this material does not reduce the efficacy of the sound conduction.

As regards the operation, complicated techniques that require too much intrinsic procedure do not become popular. In this regard, our method is easy enough to carry out without special advanced techniques and it does not require a lengthy operation.

We achieved high success rates of hearing improvement by the use of this new technique for stabilization of columella in type III and IV tympanoplasty. These results are much better than those in earlier reports [1,2]. This may indicate that the dislocation of columella without any support occurs more frequently in cases with a subtle prosthetic displacement from the best hearing position. How-

ever, our observation period (3.5 years) is not very long. As a temporal fascia string may gradually assimilate with the surrounding tissues for a long time, some force might work to move the columella in an undesirable direction and might induce dislocation of the columella. A longer period of observation is needed.

In 11 of 13 patients whose postoperative hearing was not improved within 20 dB, the causes of failure have not been found except for one patient. These causes of failure need to be elucidated in further studies.

Conclusion

The stabilization technique for columella using trimmed autologous temporal fascia is simple, easy, and cost-effective, and effective for improvement of postoperative hearing in type III and IV tympanoplasty.

References

- [1] Nichols SW. Long-term results using ossicular grafts. *Am J Otol* 1996;17:386-92.
- [2] Merchant SN, Ravicz ME, Voss SE, Peake WT, Rosowski JJ. Toynebe Memorial Lecture 1997. Middle ear mechanics in normal, diseased and reconstructed ears. *J Laryngol Otol* 1998;112:715-31.
- [3] Dogru H, Uygur K, Tuz M. Stabilization of autograft ossicles in type III tympanoplasty. *Laryngoscope* 2001;111:1500.
- [4] Torizuka T, Hayakawa K, Satoh Y, Tanaka F, Okuno Y, Maeda M, et al. Evaluation of high-resolution CT after tympanoplasty. *J Comput Assist Tomogr* 1992;16:779-83.
- [5] Wehrs RE. Hydroxylapatite implants for otologic surgery. *Otolaryngol Clin North Am* 1995;28:273-86.
- [6] Dalchow CV, Grun D, Stupp HF. Reconstruction of the ossicular chain with titanium implants. *Otolaryngol Head Neck Surg* 2001;125:628-30.
- [7] Babighian G. Bioactive ceramics versus proplast implants in ossiculoplasty. *Am J Otol* 1985;6:285-90.

論 説

側頭骨を用いた耳科手術トレーニングシステム

伊 藤 壽 一

Otosurgical Training Using Human Temporal Bone

Juichi Ito

(Kyoto University)

It is very difficult especially for young residents to master otologic surgery because of the anatomically complex temporal bone. Training by a skilled surgeon is necessary, but training using human temporal bones is most important. Only doctors who have performed repeated use of temporal bone dissection become skilled and knowledgeable in temporal bone anatomy and will have the confidence to complete actual otosurgery.

This paper will introduce the concept of surgical dissection of the temporal bone; as well as discuss of the some problems using human temporal bones.

Key words : training, otologic surgery, temporal bone dissection

はじめに

耳科手術は耳鼻咽喉科・頭頸部外科領域の手術のなかで習得に非常に時間がかかり、また困難なものである。従来、耳科手術の習得には熟練した術者の助手を務めたり手術ビデオをみたりして徐々にその技術を向上させるという方法が取られてきた。耳科手術など、顕微鏡を使用する手術と他の手術との決定的な違いは、手術は術者一人に委ねられるということであり、モニターを使っても上級者が同じ術野で直接指導するのが困難な点にある。

耳科手術のカバーすべき領域は、側頭骨内に位置し、術者はCTなどの画像をみながら各部位の3次元立体構築をしながら手術を進めていくが、これはなかなか至難の業である。耳科手術のなかでも中耳を扱う機会は比較的多いが、内耳、内耳道となると限られた人のみが正確にその空間的位置関係を把握できていることになる。蝸牛の回転がどのようになっているのか、前庭器の球形囊

斑、卵形囊斑がどこにあり、どのような位置関係になっているのか、半規管膨大部がどこにあるのか、顔面神経の走行、特に迷路部の走行はどうなっているのかは平面的な教科書の解説のみでは把握するのは困難である。さらに内耳道はどこにありどのような方向に向かっているのか、各神経が内耳道にどのように入り込んでいるのか、中頭蓋方向、後頭蓋方向からみた内耳道、蝸牛、半規管の位置はどこにあるのか、これらの問いに正確に答えられる人は非常に少ないと思われる。

耳科手術を習得するためには、まず中耳の構造の正確な把握が不可欠であることはいうまでもない。しかし、内耳、内耳道などの解剖の把握ができてさらに中耳手術手技が向上し、また新しい手術の開発につながると考えられる。欧米では耳科手術者のなかには、中耳だけでなく、内耳・内耳道の手術のエキスパートは必ずしも少なくない。しかし、わが国ではごく限られた耳科手術者のみがこのような手術を手がけているのが現状である。こ

これは手術のトレーニング法に決定的な違いがあるためと思われる。すなわち、欧米の優れた大学、病院では、ヒト側頭骨 (cadaver) を用いて、実際の手術と同様の設備を備えたトレーニングルームを有し、そこでそれらを何度も削開し、解剖学的部位を把握してから実際の手術を行い、それで疑問点が生じると再び側頭骨を使ってその疑問点を確かめるということを繰り返し行っている。また、数日から1週間近くの側頭骨 (cadaver) を用いてのトレーニングコースを設け、それに何回か参加し、実習することによって知識の習得と技術の向上を目指している。当然耳鼻咽喉科・頭頸部外科のレジデント教育プログラムにも取り入れられており、一回1000ドルから3000ドルのコースに数回参加することを義務付けている施設も多い。一方、わが国ではこのような設備を設け、プログラムを持っている施設は皆無であるのが現状である。

本稿では京都大学耳鼻咽喉科・頭頸部外科学の側頭骨実習室とヒト側頭骨 (cadaver) を用いた耳科手術実習プログラムを紹介するとともに、これからの耳科手術トレーニングシステムのありかたについて言及したい。

側頭骨実習マニュアル

側頭骨実習を行うためには優れた実習マニュアルが不可欠である。欧米では各施設で優れた側頭骨実習マニュアルを作成しそれによって実習を行っている¹⁾。多くの実習書は図を多用し、よく工夫されているが、内耳より中枢部の記載がやや不十分なものや必ずしも実際の手術を想定していないものなどもある。われわれは独自の側頭骨実習マニュアルを作成しそれに沿って実習を行っている。以下、われわれの作成した実習書を紹介する。

まず、側頭骨実習マニュアルの著者である高木明、辻純氏の実習マニュアル²⁾に記された序文を以下に転載し、当実習マニュアルの基本的概念を述べる。

(序文)

耳の手術の習熟のためには内耳を含めた側頭骨の形態を知ることが不可欠である。ところが一般臨床における耳の手術においては内耳形態の全貌、顔面神経の内耳道までの全走行などを知る機会ほとんどない。そのため、初学者はこれらのみえない構造物の障害をおそれて、十分な中耳操作ができなかったり、手術時間を浪費したりする。欧米では耳鼻咽喉科専攻医は temporal bone dissection の実習を行ったうえで臨床に臨むのが通常となっているが、わが国ではまだその整備は遅れている。

ここで改めて側頭骨実習の必要性を考えてみると次のような点が挙げられる。

1. 側頭骨の解剖が非常に複雑であって講義、書物では伝えることのできない立体構造を有し、結局自らの手で削らなければ3次元的深さを理解できない。
2. 側頭骨削開の技術がそのまま難聴、中耳炎、めまい、顔面神経麻痺などの手術的治療につながる。
3. 内耳の立体構造が機能と密接に絡んでいるので、前庭機能、難聴の理解にはその構造を知ってはじめて病態の理解が可能となる。
4. 新しい手術法、approach を考える基礎となる。
5. misorientation による致命的な事故防止に役立つ。

幸い、京都大学に本格的な側頭骨実習室が整備され、側頭骨 (cadaver) を用いた側頭骨実習コースが専門医認定前後の医師を対象に持つことが可能となった。その実習でのめざすところは次のような点である。

1) 側頭骨全体の立体構造の学習

具体的には中耳、内耳の構造、顔面神経の走行、内耳道の位置などの理解が目的となる。実習ではこれらの構造物を実際の臨床では不可能ないろいろな視点から観察できることにより、より3次元構造の体得をめざす。

2) 内耳を知って中耳手術の限界を知る

中耳手術は常に内耳、顔面神経の損傷の危険性を念頭に置きながら実施される。しかも、これらの構造物は骨に埋没して実際にはみえないので正確な空間認識が要求される。その認識を正確にするために、ふだんは削ってはならない内耳、顔面神経部を取って削り出して、その見えざる空間位置を確認する。

3) 手術手技の習熟・向上

実際の手術で顕微鏡を用いて側頭骨の入り組んだ部位を的確にみるためには一定の技術を要する。顕微鏡の適切な倍率、レンズの振り方、ドリル、吸引の使い方の基本を習熟する。

4) 機能と構造の把握

いうまでもないが、機能と構造は不可分である。半規管、耳石器の構造、空間位置を知れば、BPPV の発症機序、耳石置換法の理解が容易となる。また、外リンパ瘻が仮に正門窓の部位に起こったとすれば、蝸牛症状が全面にでるであろうなどの予想がたつ。

5) 新しい手術法の考按

singular neurectomy, cochleosacculotomy などは解剖を熟知した上で考案される手技である。また、人工内耳手

術に際して、基底回転が骨化している場合、第2回転、あるいは前庭階からの挿入となるがこれも、蝸牛の空間位置の深い理解がなければ、不可能である。

ただし、側頭骨実習に供される標本と実際の生体での手術とではかなりの差異があるので手術手技の実習として以下のような限界がある。

1. 結合織の感触が実際の手術と異なる
2. 出血がなく、骨の透明度が違う
3. 現実的でない体位で削開ができてしまう
4. 再建手術ができない
5. 疾患耳ではない

具体的には再建を伴う鼓室形成術、微妙な感触が大切なアブミ骨手術などの体得は難しい。

最近の医療を取り巻く環境をみると、耳科手術に際しても系統的な習練を積んだ後、手術に臨むことが要求されている。ぶっつけ本番は許されない時代である。厚生労働省の施策によっても耳科手術を日常的に行う施設はますます限定されたものとなり、専門医をめざす多くの耳鼻咽喉科医にとっての耳科手術の習練の場が限られつつある。そのような現状のなかで耳鼻咽喉科医の技術を高い水準に保つためには側頭骨実習をすべての耳鼻咽喉科医に必須のカリキュラムと位置づける必要がある。ただし、その維持管理のコストと人的労力は多大であるので、将来は学会主導型のよりシステム化された実習が広く行われることを期待したい。

6) 実習参加者へ

序文に書かれたようなことを念頭において実習を行うが、側頭骨 (cadaver) は貴重なものであるので余すことなく、なめ尽くすように利用、削って頂きたい。削るスピードはマイペースでよいので一つ一つのステップを確認、納得しながら進めてほしい。日常臨床では常に急がされながらの手術となるが、実習では道草を食いながらでもよい。手術書に記載がないものがみえたりすることを楽しんでほしい。内耳に穴が開いてもよい。ただし、すぐに気づく程度の知識と慎重さは求められる。そして、予期せぬことが起これば、なぜ、予期できなかったかを考えてほしい。次回には予期せぬこととならない方策を考えてほしい。判らないときは遠慮なく指導者に質問してほしい。また、各ステップをスケッチなり写真なりで記録に留めてほしい。今はデジカメで接眼レンズから容易に写真を撮ることができる。

人間の立体把握能力は意外に貧弱で、みる方向が違え

ばとたんに判らなくなる。それゆえ、側頭骨構造物をできるだけ、いろんな方向からみてほしい。機会があれば、別のアプローチによる dissection も試みてほしい。骨に埋もれた構造物を同定するにあたり、なんとなく、この辺かなといった削り方をするのではなく、現在確認できる構造物を指標として同定できるように努めてほしい。一通り dissection が形になったとしても、そのときの技量、興味のあり方でみえるものが毎回違う。その点で実習には終着点はない。今後とも許される範囲で積極的な参加をめざしてほしい。

側頭骨実習室

ヒト側頭骨 (cadaver) を用いての耳科手術トレーニングは、顕微鏡を含め、実際の手術と同じといわないまでも、手術器具も充実したものである必要がある。これは本トレーニングが単に側頭骨の解剖を熟知するのみならず、実際の耳科手術を想定して顕微鏡の操作、耳科手術用の特殊器具に慣れることをも目的とするからである。図1は京都大学の側頭骨実習室を示す。

1つのブースには顕微鏡とドリルおよび耳科手術器具を揃える。このブースを6台設置し、6人が同時に実習を行えるようにする。手術用顕微鏡にはそれぞれにCCDカメラをつけ、液晶モニターを装備した。実習中に指導者が観察でき非常に有用である。ビデオレコーダーも各テーブルに設け各自の実習内容を記録し、後の検討・反省材料とする。指導者用のブースのモニターは天井より懸垂型とし、見学者全員が観察できるようにする。なおこのモニターは大学病院中央手術室と画像音声とも接続しており、適宜ライブ手術を受信し術者と討論することができる。また、これらの映像は隣接したセミナー室とも接続しており、セミナー室で手術室、実習室を同時に中継しながら多くの人々が討論できるよう設計されている。

実習にはホルマリン固定の試料を使うことが多いので(必要に応じ冷凍の側頭骨を使用することもある)十分な換気システムを有する実習室が望ましい。なお、側頭骨の固定には一般には3点固定のポールタイプのものを使用することが多いが、比較的小さな側頭骨の保持しかできないため、われわれは加工用のバイスを利用しかなり大きい試料も固定可能なものとした。吸引装置は机の下に設置し、顕微鏡に振動が伝わりにくくした。

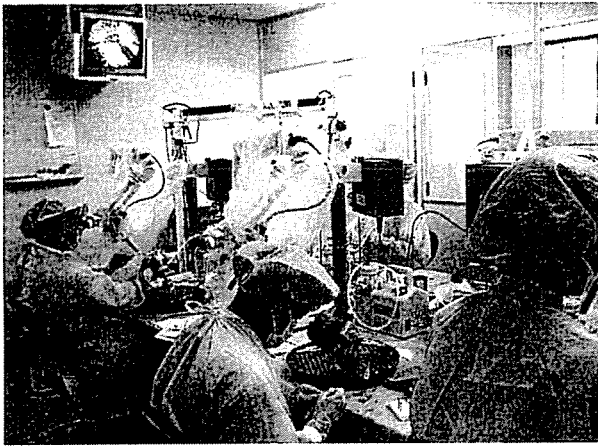


図1 京都大学の側頭骨実習室
1つのブースには顕微鏡とドリルおよび耳科手術器具を揃える。このブースを6台設置する。

側頭骨実習の手順

以下実際の実習手順とそれぞれの簡単な説明を記す。

1) 乳突蜂巣削開 (mastoidectomy)

開始位置は suprameatal spine (Henle's spine) で、頭蓋底、S 状静脈洞、外耳道を1つの3角形として削開する。乳突洞に入ったら、内側壁の外側半規管隆起を同定する。乳突蜂巣の内側壁を性状をよく観察し、骨密度が高く硬い外側半規管隆起を同定する。次にキヌタ骨を探すために、zygomatic root 部の削開を進めキヌタ骨を同定する。キヌタ骨が同定できれば一応乳突洞削開が終了したとみなす。

2) 乳突洞開放

3) 経乳突洞的上鼓室開放 (atticotomy)

4) 後鼓室開放 (posterior tympanotomy)

posterior tympanotomy は顔面神経垂直部(鼓室部)、鼓索神経、fossa incudis に囲まれた三角形を削開し鼓室に入る。慢性中耳炎、真珠腫、人工内耳手術に必要な手技である。

posterior tympanotomy の進行方向は、前方にずれると外耳道への穿孔、鼓膜輪の破損をきたす。後方にずれると顔面神経を損傷する。

5) 蝸牛開窓 (cochleostomy)

人工内耳電極挿入術を想定し、正円窓の前下方を1mmバーで削開し蝸牛基底回転鼓室階を開放する。正円窓の同定には卵円窓、Jacobson's nerve が参考になる。人工内耳電極は正円窓から挿入すると、基底回転の hook portion

で屈曲するため、より頂回転側に cochleostomy を行いまっすぐに挿入できるようにする。

6) 上鼓室開放のオリエンテーション

上鼓室開放は臨床的に弛緩部型の真珠腫の除去時に多く行われる。また耳小骨の点検、固着の有無の確認などにも重要な手技である。上鼓室への到達方法は、経外耳道的に上鼓室外側壁を開放する方法と、後方から経乳突洞的に到達する方法がある。

7) 顔面神経減荷術

顔面神経管の開放は、水平部、垂直部のみならず、中枢側は膝神経節から末梢側は顎二腹筋がみえるまで行う。

8) 外耳道後壁切除 (canal wall down)

9) 内リンパ嚢開放

内リンパ嚢は、外側半規管上に引いた Donaldson line より下方に存在するとされる。

10) 半規管の解剖

11) 前庭器の解剖

12) 卵形嚢斑、球形嚢斑の観察

外側半規管と顔面神経水平部を切除して、前庭を開放する。白色で、柔らかく、水平に前庭内に突出する utricular macula (卵形嚢斑) と、それに垂直で、骨壁に付着する saccular macula (球形嚢斑) がみえる。アブミ骨は蝸牛の前庭窓についており、その底板は卵形嚢斑の裏面の方を向いている。

13) 半規管膨大部の開放

前庭の開放を進め、半規管膨大部を観察。後半規管膨大部は顔面神経の内側に位置する。labyrinthectomy (迷路摘出術) などは、すべての感覚上皮の摘出を要する場合がある。このとき顔面神経の損傷に特に注意を要する。

14) 蝸牛管の解剖

卵円窓、正円窓の間の骨を除去する。蝸牛基底膜は、正円窓近くで下方に屈曲しており、hook portion と呼ばれる。ここでの削開時、蝸牛基底回転の終末の hook portion を保存し、正円窓との位置関係を理解することは、人工内耳手術の解剖学的位置の理解に非常に有用である。正円窓膜も一部だけでも極力保存する。蝸牛管を頂回転方向に削開すると、osseous spiral lamina (骨ラセン板)、基底膜が確認できる。正円窓膜をできるだけ残しておいたほうが、オリエンテーションの助けになる。第2回転、頂回転を開放する。第2回転の開窓はサジ状突起の下方を削開する。

| 開始 | 時間 | グループ A | グループ B |
|-------|----|---|---|
| 8:00 | 1 | registration, 挨拶, 紹介 朝食セミナー・dissection 講義 | |
| 9:00 | 3 | 実習室オリエンテーション | |
| 9:15 | | 実習 1・中耳1 mastoidectomy, posterior tympanotomy cochleostomy | dissection 講義(中耳・内耳) 側頭骨 surface anatomy ライブ手術(手術室と中継) |
| 12:00 | 1 | 昼食セミナー・(教室形成など) | |
| 13:00 | 3 | dissection 講義(中耳・内耳) 側頭骨 surface anatomy ライブ手術(手術室と中継) | 実習 1・中耳1 mastoidectomy, posterior tympanotomy cochleostomy |
| 16:00 | 2 | 反省会および歓迎パーティー | |
| 18:00 | | 解散 | |

図 2 側頭骨解剖実習コースの一例

公開コースは 4 日間で行う。図はその 1 日目を示す。

15) 内耳道の解剖

前庭から内耳道を開放する方法は、聴神経腫瘍の translabyrinthine approach として行われる。

この方法は聴覚を犠牲にするが、小脳の圧排が不要であること、顔面神経の機能を保存しやすいこと、前下小脳動脈に対するリスクが少ないことから安全性が高い。臨床の場面では、実用聴力が残存し、腫瘍が内耳道内であれば中頭蓋窩アプローチを、聴力残存がない場合は translabyrinthine approach が選択されることが多い。

なお、図 2 に公開で行う側頭骨実習プログラムの 1 例を掲載する。

側頭骨実習の問題点

上記に述べたような側頭骨実習の必要性はいうまでもないが、実習室の整備には当然費用がかかる。しかし、最も大きな問題点はヒト側頭骨 (cadaver) の入手の問題である。限られた数であれば各大学の解剖学教室、病理学教室の協力のもとに、側頭骨を使用する方法があり、実際このような方法で実習を行う場合もある。しかし、それでは実際必要とする数に対して大きく不足する。京都大学では海外から輸入したヒト側頭骨 (cadaver) を使用する場合が多いが、かなり費用がかかる (1 個の側頭骨で 1000 ドル位必要な場合もある)。さらに輸入元の米国でも多くの施設でヒト側頭骨 (cadaver) を使用するた

め、絶対数が不足する傾向にある。ヒト側頭骨 (cadaver) の不足を補い、側頭骨実習を行う手段として、人工材料を用い、側頭骨 CT から再構築した人造側頭骨を使用して基本的な解剖実習を行う方法も考案されている。また、コンピューター画面上に側頭骨を再現し手術のシミュレーションを行う方法もあり、これらの精度が高まれば今後利用する価値はあると思われる。

一方、このような実習に対する倫理的、法的問題、さらには輸入側頭骨に含まれる化学物質の問題³⁴⁾もある。倫理的問題としては解剖実習用の献体された屍体がある一方で海外から輸入の側頭骨を利用することに対する批判である。このような指摘はある意味ではもったもであるが、上記のように実際必要とされている数に対し絶対的に不足している現実では輸入のものを使用するのはやむをえないのではないと思われる。また、解剖実習はどんな小さなものであれ、指定された解剖学実習室で行わなくてはならないという法的問題もこれから解決しなくてはならない問題であろう。

今後の方向性

現在行われているような実習は、費用の問題、側頭骨入手が困難な点、倫理的な問題もあるが、耳科手術のスキルアップ、医療事故などを避けるためにも必須であり、少数の施設だけでなくできるだけ多くの施設で行う

べきであろうと思われる。

このような実習をさらに充実させるためには、実際の手術を想定して、事前に側頭骨のCTを撮影し、実習時にCTとの比較検討を行うことや、新しい耳科手術機器も実習に取り入れるなども必要であろう。逆に、耳科手術に新たな機器が導入される場合はヒト側頭骨(cadaver)を用いこれらの機器の精度を確かめることも必要であろう。具体的には、ナビゲーションシステム、内視鏡の使用、将来のロボット手術の応用の際にこれらの機器をヒト側頭骨(cadaver)で確かめることも有用と思われる。また文中にも述べたがヒト側頭骨(cadaver)が不足する現状では、使用できる部分は人造側頭骨を使用し、またコンピューターによるバーチャルな画面での実習シミュレーションも有用であろう。

最後に、耳科関連学会などもこのような実習に対して

認定制度を作り、各実習に対しバックアップ体制を取ることが望ましいと思われる。

本号カラー図説に辻、高木氏著の「側頭骨手術解剖実習」が掲載されているので、参照されたい。

参考文献

- 1) Nelson RA : Temporal bone anatomy.
- 2) 高木 明, 辻 純 : 側頭骨臨床解剖実習.
- 3) 村上 弦 : 臨床解剖研究と正常解剖体業務上の制約. 日本医事新報 4001 : 24 ~ 29, 2000.
- 4) 村上 弦 : 外科系医師の卒後研修としての fresh cadaver dissection と生前同意. 日本医事新報 4226 : 20 ~ 25, 2005.

別刷請求先：伊藤壽一

〒606-8507 京都市左京区聖護院川原町54

京都大学大学院医学研究科耳鼻咽喉科・頭頸部外科学