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CT Imaging of the Eustachian Tube Using Focal Contrast Medium Administration: A Feasibility Study

Benedicte Falkenberg-Jensen, MD; Juha Silvola, MD, PhD; Helene Laurvik, MD; Andreas Lervik, DVM, Dipl. ECVVA; Joanna Fenn Kristiansen, MSc; Greg Jablonski, MD, PhD; Einar Hopp, MD, PhD

Objectives: We aim to develop an imaging technique for visualization of the Eustachian tube (ET) lumen.

Study Design: A prospective, experimental study in an animal model and in human cadaver specimens.

Methods: Applying iodixanol to the middle ear in two human temporal bone specimens, followed by computed tomography (CT) examinations, we optimized contrast dilution, CT algorithm, and head positioning for visualization of contrast passage through the ET.

Myringotomy was performed on eight rabbits. Based on the cadaver study, a 20% iodixanol solution was applied to the middle ear, and subsequent CT scans were performed to observe iodixanol in the epipharynx. For some animals, the procedure was repeated on the contralateral ear. We performed the procedure twice on four subjects. Twenty examinations were included.

Iodixanol appearance in the ET and the epipharyngeal orifice was assessed qualitatively on CT scans. The tympanic membrane was inspected after 1 or 2 weeks, and histopathological examination of six contrast-exposed temporal bones was performed.

Results: The cadaver study provided information on imaging technique and contrast dosage. In rabbits, iodixanol passed through the ET in 19 of the 20 ears. Qualitatively, optimal visualization was seen after 9 to 12 minutes. Clinical inspection after 1 or 2 weeks revealed normal middle ear status. Histopathological samples showed no sign of inflammatory reaction in the tympanic membrane, middle ear, or ET.

Conclusion: Iodixanol application to the middle ear is feasible, safe, and demonstrates patency of the ET.

Key Words: Eustachian tube, contrast medium, CT, balloon dilation, rabbit.

Level of Evidence: N/A.

INTRODUCTION

Middle ear ventilation problems are connected to infectious, inflammatory, or even destructive disease of the middle ear (ME).[1] Dysfunction of the Eustachian tube (ET) is considered to be one of the main reasons for ME ventilation problems.

The ET consists of a bony part leading from the tympanic cavity and connecting to a cartilaginous part, which is considered the functional part and leads to the epipharynx. The point of connection between the two parts, the isthmus, is anatomically the narrowest portion. However, pathological processes such as inflammation, tissue hyperplasia, and malignancies can lead to stenosis along the lumen. The ET's explicit manner of function is still discussed, but studies have shown an opening of a more peristaltic character rather than a simultaneous opening of the entire lumen.[2] Muscles levator veli palatini, tensor veli palatini, and the medial pterygoid are all involved in the opening process. Because the lumen is only open one portion at a time in healthy subjects, diagnostic radiologic imaging is challenging. With computed tomography (CT) imaging, soft tissue is homogeneously gray. Magnetic resonance imaging (MRI) may define muscle and cartilage of the ET. Both on CT and MR images, sporadic air can be seen along the expected course of the ET's cartilaginous portion, but most of the lumen is closed. Thus, the lumen is not visualized and the ET's patency cannot be evaluated. As Smith et al. conclude,[3] there is a need for refined imaging methods.

In recent years, balloon catheter dilation of the cartilaginous part of the ET has emerged as a treatment option with beneficial effect on ET dysfunction.[4–7] According to Schröder et al., balloon dilation has a positive effect on ET function in more than 70% of cases.[6] Nevertheless, it is a challenge for the otologist to select

From the Department of Radiology and Nuclear Medicine (B.F-J., J.F.K., E.H.); the Department of Otorhinolaryngology, Head and Neck Surgery (J.S., G.J.); the Department of Pathology (H.L.), Oslo University Hospital, Rikshospitalet; the University of Oslo (B.F-J., G.J.); and the Norwegian University of Life Sciences (A.L.), Oslo, Norway.

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Send correspondence to Benedicte Falkenberg-Jensen, MD, Department of Radiology and Nuclear Medicine, Oslo University Hospital, Rikshospitalet, Postboks 4950 Nydalen, 0424 Oslo, Norway. E-mail: bfalke@ous-hf.no

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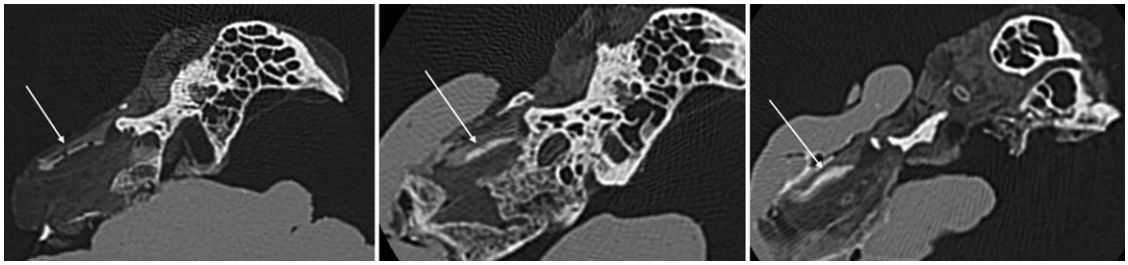

Fig. 1. The cartilaginous part of the Eustachian tube in human cadaver specimen filled with 5%, 15%, and 20% iodixanol from left to right. Arrows: Contrast medium in the cartilaginous part of the Eustachian tube.

patients for the treatment, not knowing whether the pathology is in the cartilaginous part of the ET. At present, predilation CT is performed to reveal any anatomical variants contraindicating dilation, or on rare occasions to reveal an epipharyngeal mass causing compression of the ET. Imaging has not yet contributed to existing knowledge regarding the functional cause of obstruction or the obstruction level. The question remains whether preoperative selection could be improved if the level and origin of pathology was known. This would require visualization of the ET lumen on radiologic images.

Our main hypothesis is that contrast medium (CM) applied to the ME will drain into the ET and be visible in a subsequent CT examination of the temporal bone and epipharynx, providing clinically important information before ET balloon dilation.

Although studies using CM to validate the function of the ET have been performed,[8–12] no systematic studies have been conducted. Before performing studies on humans, we needed to assess possible negative effects of the CM to determine a CM dilution that would be distinguishable from both bone and soft tissue, and to confirm that CM does pass from the ME to the epipharynx in assumed healthy individuals.

To address all of these aspects, we conducted an animal trial with the opportunity to perform multiple CT examinations. The project had the following objectives: 1) To determine the ideal parameters regarding contrast dilution, CT algorithm, and head positioning for visualization of the ME anatomy and contrast passage through the ET; and 2) to explore the feasibility and safety of CM injection into the ME.

To achieve this, we combined two methods in our study design. The first objective was assessed through studies on human temporal bone cadaver specimens and the second objective through a study on live rabbits. Sucheston has described the ET in rabbit as similar to the human ET on both a gross and microscopic level.[13] We believe the method to be applicable to humans with tympanostomy tubes. The animal study was approved by the national animal research authority and was conducted according to European Communities Council Directive of November 24, 1986 (86/609/EEC) and the guidelines of Animal Research: Reporting In Vivo Experiments.

MATERIALS AND METHODS

Two human temporal bone cadaver specimens that had been preserved by freezing were used. In the initial preparation, the auricles were removed; in one specimen, part of the ET's epipharyngeal orifice was missing. Iodixanol (Visipaque, GE Healthcare, Oslo, Norway) was chosen as CM due to its isoosmolar properties. Dilutions of 5%, 10%, 15%, 20%, and 25% were made in advance by using NaCl 9 mg/mL (B. Braun Melsungen AG, Melsungen, Germany) as a dilutant. To avoid movement, the temporal bone specimens were fixed to a plastic tray with Play-Doh (Hasbro Inc., Pawtucket, RI) in what would have been an oblique, decubitus position. Aided by otomicroscopy, an experienced otologist performed the myringotomy. Starting with the lowest iodine concentration, contrast medium was applied to the ME, succeeded by two CT (Aquilion One, Toshiba, Minato, Japan) scans employing a constant mA of 200: one at 120 (kilovolt) kV and one at 135 kV. Following scan acquisition, the ME cavity was flushed with saline (NaCl 9 mg/mL), and the procedure repeated with each contrast dilution in turn. Images were reconstructed with both 8-cm and 16-cm display field of view (DFOV).

Two experienced radiologists subjectively evaluated image quality with respect to the optimal contrast medium dilution and the optimal spatial and contrast resolution. The contrast dilution that was most easily distinguishable from soft tissue without masking bone tissue was deemed the best choice (Figure 1).

Ten rabbits (New Zealand White, female) were purchased through the Centre for Comparative Medicine at Charles River Laboratories Inc. (Châtillon-sur-Chalaronne, France) and were kept in individual cages in a separate room in the Centre for Comparative Medicine. All animals had identification ear tattoos in one ear and in addition were given an individual letter (A–J). Each cage was marked with both identifiers. Before the experiments started, the animals were acclimatized in the Centre for Comparative Medicine (Charles River Laboratories Inc.) for 2 weeks. Every animal had a healthy appearance and a good appetite during this period.

All animals had access to food and water until premedication was given. Premedication consisted of fentanyl and Fluanisone (Hypnorm, VetaPharma, Leeds, UK). The animals were instrumented with an intravenous catheter in each auricular vein. Anesthesia was induced with a slow intravenous injection of propofol (Propofol-Lipuro; B. Braun Melsungen AG) and maintained by propofol infusion at a constant rate using a syringe driver. Dexmedetomidine (Dexdomitor, Orion Corporation, Turku, Finland) was administered as a constant rate infusion using a syringe driver. All animals were monitored for signs of inadequate anesthetic depth, which included an increase in pulse rate, respiratory rate, and spontaneous movement in response to stimulation. Arterial oxygen saturation of hemoglobin and pulse rate were monitored using a pulse oximeter.

The tympanic membrane was visualized through a 3-mm examination tube. Lidocaine (Lidokain 10 mg/mL, PharmaPlus,

TABLE I.
Showing Animals A–J, the Number of Contrast-Enhanced CT Examinations (Procedures) Conducted on Each Animal, the Number of Successful CM Passages, and Number of Ears Excluded.

	No. Procedures	No. Contrast Medium Passages to Epipharynx	No. Ears Examined With CT Without Contrast	No. Ears Excluded	No. Ears Sent to Histopathology
A	2	2	2	–	2
B	2	2	–	–	–
C	2	2	2	–	2
D	4	4	–	–	–
E	4	2	–	2	–
F	2	1	2	1	2
G	4	3	–	–	–
H	3	3	–	–	–
I	–	–	1	–	2
J	–	–	1	–	2
Total	23	19	8	3	10

CM = contrast medium; CT = computed tomography.

Oslo, Norway) was administered locally into the distal auricular canal. Aided by otomicroscopy, an experienced otologist perforated the tympanic membrane with a 22-G needle attached to a syringe with contrast dilution by a flexible connection tube with Luer lock ends. The CM was slowly injected into the ME until achieving a visual impression of the ME being full but not expanded. The injected volume was 0.3 to 0.4 mL for all animals.

With the animal in a lateral decubitus position, CT scans of the temporal bone and epipharynx were performed with 200 mA, 120 kV, and 10.3 cm DFOV at 3-minute intervals for 12 minutes. After the experimental procedure, propofol, dexmedetomidine, and oxygen delivery were discontinued. Monitoring continued until the rabbits were sitting in sternal recumbency.

The first rabbit (A) was given a 15% CM solution in its left ear based on the results of the human cadaver study. However, the contrast medium appeared less dense than desired. The concentration was therefore increased to 20% for the remaining ears. In spite of the increased CM concentration, bone was easily distinguishable.

The animals were followed for 1 or 2 weeks to explore whether the procedure induced inflammation. Before euthanasia, all animals underwent a noncontrast CT scan, and the CM application was repeated and new CT scans acquired for most of the subjects.

In total, the eight animals underwent the CM procedure 23 times, with a maximum of two procedures per ear (Table I).

Euthanasia was performed in continuity with the final sedation by an intravenous injection of 20-mL potassium chloride (1 mmol/mL; Kaliumklorid B. Braun, B. Braun Melsungen AG) or by blood drainage through the jugular vein.

The two remaining animals (I and J) had not undergone the procedure and were euthanized on the last day of the experiments to serve as control animals in the histopathological evaluation. In total, three of the animals given CM and the two controls were dissected for histopathological purposes after euthanasia. The latter two underwent a CT examination of the temporal bones (without myringotomy and CM injection) before euthanasia to exclude any ME effusion.

After the animals were sacrificed, the left and right temporal bones and surrounding tissue were dissected out in one piece. After fixation in 10% buffered formalin, frontal slices were cut by a diamond saw and placed in separate cassettes to decalcify for 36 hours. The decalcification time was kept as short as possible to prevent damage to the morphology in the final microscopic sections. Thirty-six hours proved to be enough time to soften the tissue. The specimens were then paraffin-embedded, and microscopic sections were cut from the paraffin blocks and stained with hematoxylin and eosin.

RESULTS

In the cadaver experiment, 15% iodixanol had a density lower than bone but was easily distinguishable from soft tissue (Figure 1). Twenty percent dilution also provided satisfactory visualization, whereas $< 15\%$ was too weak and $> 20\%$ was too dense to distinguish from soft tissue and bone, respectively.

Both radiologists subjectively evaluated image quality by comparing images acquired at 120 kV with 135 kV, and images reconstructed with 8 cm DFOV with 16 cm DFOV. The optimal combination was deemed to be 120 kV and 8 cm DFOV.

In total, eight rabbits (animals A–H) were in the CM group. The procedure was repeated on four of the animals after 1 or 2 weeks (Table I), resulting in a total of 23 examined ears. Of these, two ears (same animal) were excluded due to aspiration of stomach contents and subsequent pharyngeal edema (following an accidental overdose of dexmedetomidine), and one ear was excluded due to contrast from the contra lateral ear contaminating the ET orifice and ventral ET lumen. Of the 20 remaining ears, the CM passed through the ET in 19 of the cases. In some of the cases, the CM coated the ET lumen in its full length and could be visualized in a single image using thick MIP (maximum intensity projection) reconstruction.

Although CM was seen in the epipharyngeal orifice on the early series (3 and 6 minutes), the course of the ET was seen more clearly on the later series (9 and 12 minutes) (Figure 2).

The CT scans on the day of euthanasia did not reveal remnants of CM in the ME, mastoid, or ET in any of the animals.

Clinical inspection was performed 1 or 2 weeks after the first examination. All of the animals had

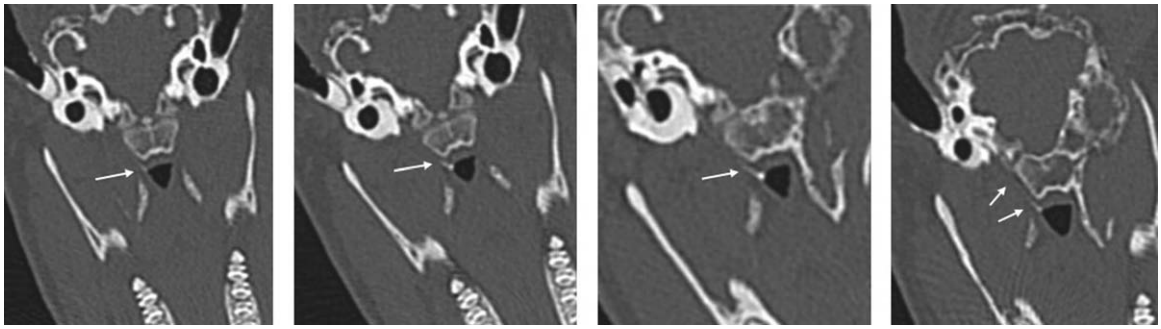

Fig. 2. Contrast filling of the Eustachian tube at different intervals after deposition in the middle ear 3, 6, 9, and 12 minutes from left to right. Arrow at the right Eustachian tube orifice in nasopharynx at the Torus tubarius.

normal ME status. One animal had the remains of a minor perforation in one eardrum and a small bulla in the other. In the rest of the animals, the perforations had healed (Figure 3).

Gross histopathological samples taken from both sides in three animals showed no sign of inflammatory reaction in the eardrum, ME, or ET. Microscopic examination of the stained slides revealed good representation of the inner part of the auditory canal and the tympanic cavity of all the ears. The tympanic membrane could be appreciated in most cases. Pathological changes were not found in any of the examined structures or the surrounding tissues (Figures 4 and 5).

The optimal contrast dilution in the animal experiment was considered to be 20% when qualitatively evaluated using the criteria described in Material and Methods.

DISCUSSION

In this study, we present a systematic approach to establish an imaging method for evaluation of ET patency by deposition of CM in the ME. The application of iodixanol in the ME is off-label. The method was proven feasible in anesthetized rabbits, and clinical and histopathological examinations revealed no adverse effects of CM.

The choice of rabbits was, in addition to size and availability, based on our knowledge on ET gross and microscopic anatomy and physiology in rabbits. Suchesston has shown that the ET in rabbits has the same gross anatomy, with the same type of cartilage, arrangement of goblet cells, and mucoserous glands as humans. Like humans, and unlike some other species, they have lymphoid tissue surrounding the ET.[13]

Regardless of animal species, the ability to monitor symptomatology is limited, and the low number of subjects is in itself a limitation. We have chosen to present each CM injection as individual experiments—not considering whether the same animal was operated twice—due to the observations that all animals were healthy and there were no signs of inflammation after the first procedure.

Sedation of the animals prevented us from knowing whether the swallowing reflex was intact and, furthermore, whether the appearance of iodixanol in the epipharynx was due to an active act of swallowing or passive drainage. This will become more evident in future human studies. These will also be the issue regarding the timing of the CM passage through the ET. Not only is the species different, but the physiological conditions will also vary because the animals were sedated. In addition, the human head is composed of more tissue, both soft and bone, than the rabbit head and the temporal bone specimens used. It is therefore

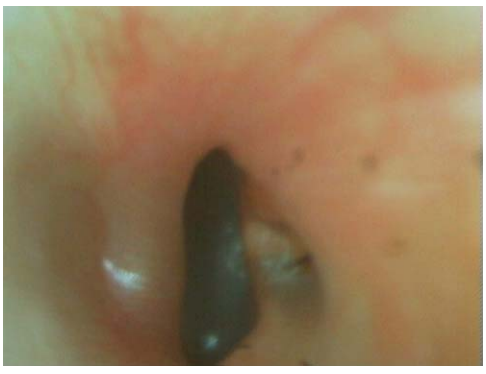

Fig. 3. Photograph taken through otoscopic microscope 2 weeks after the tympanic membrane perforation. The perforation has healed, and there are no inflammatory signs.

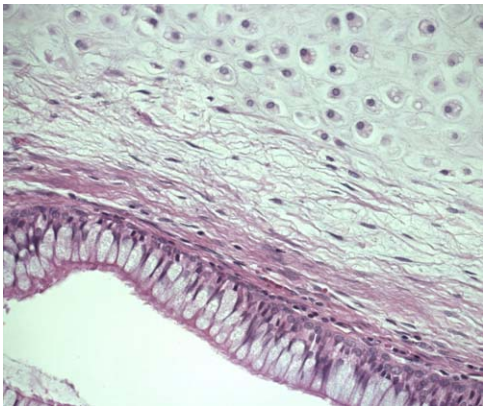

Fig. 4. Microscopy of the ciliated epithelium lining the Eustachian tube, stained with hematoxylin-eosin, from an animal twice examined with contrast medium, with no inflammatory signs.

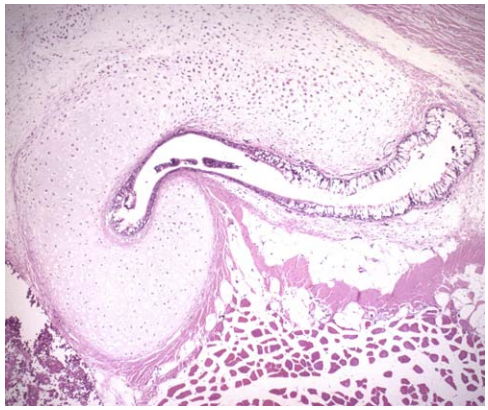

Fig. 5. Microscopy image of the cartilaginous part of the Eustachian tube, stained with hematoxylin-eosin, in contrast medium-exposed rabbit. There are no inflammatory signs.

likely that patient studies will require adjustments to the DFOV, kV, amount and degree of iodine dilution administered, and the time lapse between CM injection and CT acquisition.

In future patient studies, the CM should be deposited through a tympanic tube. Most patients with ME disease and assumed ET dysfunction already have tubes, or tubes are installed during their first visit to the otologist; therefore, unnecessary perforation of the tympanic membrane can be avoided. Long-term complication rate due to tympanic tubes is low.[14] Although the study does not predict how the method will work on patients, we believe that imaging in a lateral decubitus position also will be preferable in human studies, with the face slightly rotated toward the CT table and unilateral contrast deposition in the superiorly positioned ear, hence taking advantage of gravity.

Our study demonstrated indirectly that the ET is open by detecting CM in the epipharynx. Nevertheless, it is not given that the absence of CM in the epipharynx is synonymous with stenosis. Neither is it certain that the method will help us differentiate the normal from the pathological, or the level of pathology.

Potential hazards pertaining to the method must be addressed. Although this will be an off-label study, and the method has not been systematically tested before, we do not expect contrast-induced complications. We consider the risk of CM-induced anaphylactoid reactions to be the same as for other examinations using iodine-based CM. Iodixanol is approved and well documented for most mucosal spaces, and we assume the absorption rate through the mucous membrane to be equivalent in the ET. Given the low CM volume and dose, nephrotoxicity is also highly unlikely. Inflammatory reactions to the CM are absent in our study. Several studies have been done on the passage of gadolinium through the round window into the perilymphatic system.[15] It is possible that a similar diffusion will occur with iodine-based CMs. Nevertheless, these contrast agents are approved for intrathecal and intravenous use, systems also known to have a substance exchange with the labyrinthine system. Therefore, we consider the risk of ototoxicity to be minimal.

Finally, the radiation dose with 120 kV will be slightly lower than that of our current temporal bone CT protocol (135 kV, dose-length product 150, effective dose 0,345 mSv).

Previous studies have shown both antegrade and retrograde passage of CM through the ET.[8–12] Our study contributes knowledge regarding safety, CT parameters, and CM volume and dilution. A study of patients with chronic ME disease who have tympanic drainage tubes will be conducted based on our findings.

CONCLUSION

In rabbits, diluted iodixanol can fill and visualize the ET on CT images. Contrast medium passes from the ME to the epipharynx in 19 of 20 cases. The risk of mucosal inflammatory changes following the CM injections seems to be small.

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