

Journal of Visualized Experiments

Intrathecal application of a fluorescent dye for the identification of cerebrospinal fluid leaks in cochlear malformation

--Manuscript Draft--

Article Type:	Invited Methods Article - Author Produced Video
Manuscript Number:	JoVE60795R3
Full Title:	Intrathecal application of a fluorescent dye for the identification of cerebrospinal fluid leaks in cochlear malformation
Section/Category:	JoVE Medicine
Keywords:	cerebrospinal fluid leaks (CSF); fluorescein; temporal bone surgery
Corresponding Author:	Nora Weiss Universitätsmedizin Rostock Rostock, Mecklenburg Vorpommern GERMANY
Corresponding Author's Institution:	Universitätsmedizin Rostock
Corresponding Author E-Mail:	NoraMagdalena.Weiss@med.uni-rostock.de
Order of Authors:	Nora Weiss Ingo Andus Armin Schneider Stefanie Schröder Sebastian P Schraven Sönke Langner Robert Mlynski
Additional Information:	
Question	Response
Please indicate whether this article will be Standard Access or Open Access.	Standard Access (US\$1200)

Nora Magdalena Weiss
Department of Otorhinolaryngology – Head and Neck Surgery
University Hospital Rostock
Doberaner Straße 137-139
18055 Rostock, Germany
Email: nora-magdalena.weiss@med.uni-rostock.de

September 18, 2019

Dear Dr. Nandita Singh, dear Ladies and Gentlemen Editors,

Thank you for your kind invitation to submit our video to *JoVE*. We are pleased to submit an original authors produced video entitled

“Intrathecal application of a fluorescent dye for the identification of cerebrospinal fluid leaks in cochlear malformation”

to be considered for publication in *JoVE*.

CSF leaks are a challenging task concerning skull base surgery. A method of visualizing their origin is using intrathecal administered fluorescein. We describe this method applied in temporal bone surgery to assist the surgical orientation in cases of inner ear malformation. Our clinical case exhibits bilateral congenital ear-malformation (right ear: cochlear aplasia, left ear: Incomplete Partition Type I). We consider this case to be suitable for demonstrative purposes since it i) illustrates the scope and possibilities of the method and ii) also addresses its importance in a clinical setting.

For this reason, we believe that this video is suitable for publication in *JoVE*. It combines the detailed description of visualizing CSF leaks in conjunction with the report of a clinically challenging case.

The manuscript and the video have not been published and are not under consideration for publication elsewhere. We have no conflicts of interest to disclose.

Thank you for your consideration.

On behalf of all co-authors,

Sincerely,

Nora M. Weiss, MD
Resident and Research Fellow
Department of Otorhinolaryngology – Head and Neck Surgery, University Hospital Rostock

TITLE:

Intrathecal Application of Fluorescent Dye for Identification of Cerebrospinal Fluid Leaks in Cochlear Malformations

AUTHORS AND AFFILIATIONS:

Nora M Weiss¹, Ingo Andus², Armin Schneider³, Sönke Langner⁴, Stefanie Schröder¹, Sebastian P Schraven¹, Robert Mlynski¹

¹Department of Otorhinolaryngology, Head and Neck Surgery, Otto Koerner Rostock University Medical Center, Rostock, Germany

²Rostock University

³ARRI Medical GmbH, Türkenstraße 89, München, Germany

⁴Department of Radiology and Institute of Diagnostic and Interventional Radiology, University Medical Center Rostock, Rostock, Germany

Corresponding Author:

Nora Magdalena Weiss (nora-magdalena.weiss@med.uni-rostock.de)

Tel: +49 381 494 8301

E-mail Addresses of Co-Authors:

Ingo Andus (ingo.andus@uni-rostock.de)

Armin Schneider (aschneider@arrimedical.de)

Sönke Langner (soenke.langner@med.uni-rostock.de)

Stefanie Schröder (stefanie.schroeder@med.uni-rostock.de)

Sebastian Schraven (Sebastian.Schraven@med.uni-rostock.de)

Robert Mlynski (RobertMlynski@med.uni-rostock.de)

KEYWORDS:

cerebrospinal fluid, CSF, leaks, fluorescein, temporal bone surgery

SUMMARY:

Intrathecally applied fluorescein is used to achieve intraoperative visualization of CSF leaks. This protocol describes a lumbar puncture, the application of 5% fluorescein, and intraoperative visualization using a fully digital microscope.

ABSTRACT:

In cases of cerebrospinal fluid (CSF) leaks, reliable detection of their origins is needed to seal the leak sufficiently and prevent complications, such as meningitis. A method is presented here using intrathecal administered fluorescein in a clinical case of bilateral congenital ear malformation. A fluorescent dye is administered intrathecally to achieve intraoperative visualization of CSF leaks. The dye is applied 20 min before surgery, and concentration of 5% is used. Per every 10 kg of body weight, 0.1 mL of the fluid is applied intrathecally. The fluorescein is visualized using a fully digital microscope. The origin of the fluid leak is identified in the stapes footplate. During primary surgery, it is sealed, and cochlea implantation is performed for

hearing restoration. In this specific case, 6 weeks later, the implant was explanted due to acute meningitis, and the electrode array was left as a spacer. Postoperatively, in the aural smear, β -transferrin was detected. During a revision mastoidectomy, dislocated coverage of the leak was found. The stapes was removed and oval window sealed. Five days after revision surgery, no β -transferrin was detected in the aural smear. During the revision of cochlea implantation 6 months later, intact coverage of the oval niche was observed. Thus, intrathecal fluorescein application proves to be a reliable tool for the detection of CSF leaks. It facilitates the orientation in malformations and complicated or unknown surgical situs. In the literature, its use is described for CSF fistulas in endonasal surgery but is rarely described in skull base and mastoid surgeries. The method has been used successfully in several cases with CSF leaks, and the results confirm the feasibility of safely accessing the origin of the leak.

INTRODUCTION:

CSF leaks can be caused by trauma, preexist congenitally, or appear spontaneously. Clinically, they appear via otoliquorrhea or rhinoliquorrhea and can be confirmed by positive β -transferrin secretion^{1,2}. In cases of CSF leaks, reliable detection of its origin is needed to seal the leak sufficiently and prevent complications, such as meningitis.

Intrathecal fluorescein application (IFA) has been known to be highly sensitive in detecting CSF leaks after neurosurgical skull base operations³. However, there is no common consensus on its exact applications (e.g., concentration, amount of fluid, addition of other drugs such as dexamethasone)^{3,4}. A majority of cases describe transnasal endoscopic skull base surgery, while a standardized method of visualizing CSF leaks in the temporal bone via microscopy⁵⁻⁷ is missing. These leaks frequently occur at multiple localizations and bear the risk of recurrence if their whole extent is not visualized during surgery⁸. Specifically, a transmastoid approach only has been found to bear the risk of recurrent leaks⁸.

This protocol presents the use of intrathecal administered fluorescein via a transmastoid approach. A clinical case of bilateral congenital ear malformation (right ear: cochlear aplasia, left ear: incomplete partition type I), in which minor trauma led to perforation of the left stapes footplate in the middle ear, is also presented. A similar case has only been reported once⁹.

PROTOCOL:

This protocol was approved by the local ethics committee in accordance with the Helsinki Declaration (Reg. No. A2019-0214). Informed consent was obtained from all participants. In the presented case involving a child, written informed consent was obtained from both parents.

NOTE: Fluorescein is a fluorescent dye that emits green light (520–530 nm) when stimulated with blue light (of wavelength \sim 485 nm). It is used for visualization of the tear film in ophthalmology. Its intrathecal use is off-label and based on an individual therapeutic agreement. Fluorescein-Natrium at a 10% concentration is available and regularly used in fluorescein angiography of the ocular fundus in ophthalmology.

89
90 **1. Preparation for surgery**
91

- 92 1.1. Obtain audiological testing, high resolution computed tomography (HRCT), and
93 magnetic resonance imaging (MRI) of the skull base.
94
95 1.2. Obtain written, informed consent from the patient regarding off-label use of fluorescein.
96
97 1.3. Perform a lumbar puncture and place a lumbar drain for later application of the dye.
98
99 1.4. Use fluorescein sodium (10% concentration). For lumbar application, dilute the
100 fluorescein to 5% concentration with water.
101
102 1.5. Apply 0.1 mL of the dilution per 10 kg of body weight, with a maximum of 1 mL,
103 intrathecally.
104
105 1.6. Perform the application immediately before surgery to achieve intraoperative
106 visualization.
107
108 NOTE: When applying the method to children, the application should be performed under
109 general anesthesia.
110
111 1.7. Perform general anesthesia according to the patient's individual clinical history and risk
112 factors and according to the decision of the anesthesiologist.
113
114 1.8. Cover the microscope with a blue light filter to visualize the fluorescein, or use a fully
115 digital microscope.
116
117 1.9. Use sterile draping to cover the patient.
118

119 **2. Surgery**
120

- 121 2.1. For the transmastoid approach, perform a mastoidectomy and posterior tympanotomy.
122
123 2.2. Expand the surgery according to the expected origin of the leak.
124
125 2.3. As a first landmark, expose the dura to the middle cranial fossa.
126
127 2.4. Thin the posterior canal wall of the outer ear canal.
128
129 2.5. Expose the short incus process in the antrum and identify the lateral semicircular canal.
130
131 2.6. Expose the chorda facial angle at which the chorda tympani leaves the facial nerve.
132

2.7. Open the facial recess. Leave the facial nerve with a bony coverage.

2.8. Access the middle ear through a posterior tympanotomy. The origin of the fluid leak can easily be identified in the stapes footplate (**Figure 2**).

2.9. Use fascia from the temporal muscle and absorbable collagen fibrin sealant patches to seal the CSF leak until no more fluid leaves the footplate. The stapes suprastructure stabilizes the sealant.

REPRESENTATIVE RESULTS:

In the presented case, minor trauma led to a CSF leak from the nose in a 10 month-old child. Magnetic resonance imaging (MRI) revealed a bilateral temporal bone malformation with aplasia of the right cochlea and dilation of the left cochlea and vestibule with absent interscalar septum identical to an incomplete partition type one. Accordingly, brainstem-evoked audiometry was performed (**Figure 1**) and confirmed bilateral deafness.

Since high resolution computed tomography (HRCT) did not reveal the origin of the CSF leak, fluorescein-assisted transmastoidal otobasal exploration with simultaneous cochlear implantation was indicated. During primary surgery, the transmastoidal approach was used to identify the CSF in the stapes footplate (**Figure 2**) to seal the leak and perform cochlea implantation to restore hearing.

Six months after surgery, the child suffered from meningitis caused by mastoiditis. The implant was removed, and the electrode array was left as a spacer. Postoperatively, the aural smear detected β -transferrin¹⁰. Revision mastoidectomy was performed showing dislocated coverage of the leak in the stapes footplate. The stapes was completely removed (**Figure 3**), and the oval window was sealed using temporal muscle, temporal fascia, and absorbable collagen fibrin sealant patches. The stapes was investigated under an electron microscope, showing the malformed footplate with perforations. To reduce pressure on the leak, a lumbar puncture was performed daily. Five days after revision, no β -transferrin was detected. During cochlea implant revision 6 months later, coverage of the oval niche was intact.

FIGURE LEGENDS:

Figure 1: Audiometric results. Brainstem-evoked response audiometry (BERA) showing no reproducible brainstem responses, confirming bilateral deafness.

Figure 2: Identification of the leak in the left stapes footplate. Asterisk: horizontal semicircular canal, arrow: facial nerve.

Figure 3: Extraction of the stapes. Asterisk: lateral semicircular canal, arrow: stapes footplate, cross: eminentia pyramidalis.

Figure 4: Electron microscope investigation of the extracted stapes. Shown is the electron microscopic investigation of the extracted stapes with the congenital perforation. Scale bar indicates the magnification.

DISCUSSION:

While many reports concerning CSF leaks focus on a transnasal endoscopic approaches in skull base surgery, a standardized method of visualizing CSF leaks of the temporal bone via microscopy⁵⁻⁷ is lacking. This protocol describes an adaption of existing literature reviews for use in microscopic ear surgery.

The application of fluorescein has been described as a safe method in skull base surgery and uses endoscopic techniques. Side effects are assumed to depend on the dose¹¹. Though the use is off-label, no side effects were observed in all patients receiving intrathecal fluorescein. Furthermore, the dye can be applied repeatedly. No severe long-term side effects are typically expected¹². In the literature, intrathecal use of fluorescein is described for CSF fistulas in endonasal surgery¹³ but is rarely described in skull base and mastoid surgery. Especially in transmastoid surgery, CSF leaks may occur in a multilocular manner and tend to recur⁸. Waiving a visualization method may lead to recurrent leaks or insufficient sealing methods.

Intrathecal fluorescein is used for cases in which CSF leaks have been confirmed but cannot be visualized via CT or MRI¹³. Its intrathecal use is proved to be a reliable tool for the detection of CSF leaks^{11,12,14}. It facilitates the orientation in malformations and complicated or unknown surgical situs. This method has been used successfully in several of cases involving CSF leaks, and the results confirm the feasibility of safely accessing the origin of the leak.

The protocol does not require drastic changes in common surgical methods. The only additional effort that must be performed is the lumbar drain. We consider the advantages in visualization highly superior to the potential disadvantages concerning the small risk of a lumbar puncture.

DISCLOSURES:

The authors declare no financial disclosures.

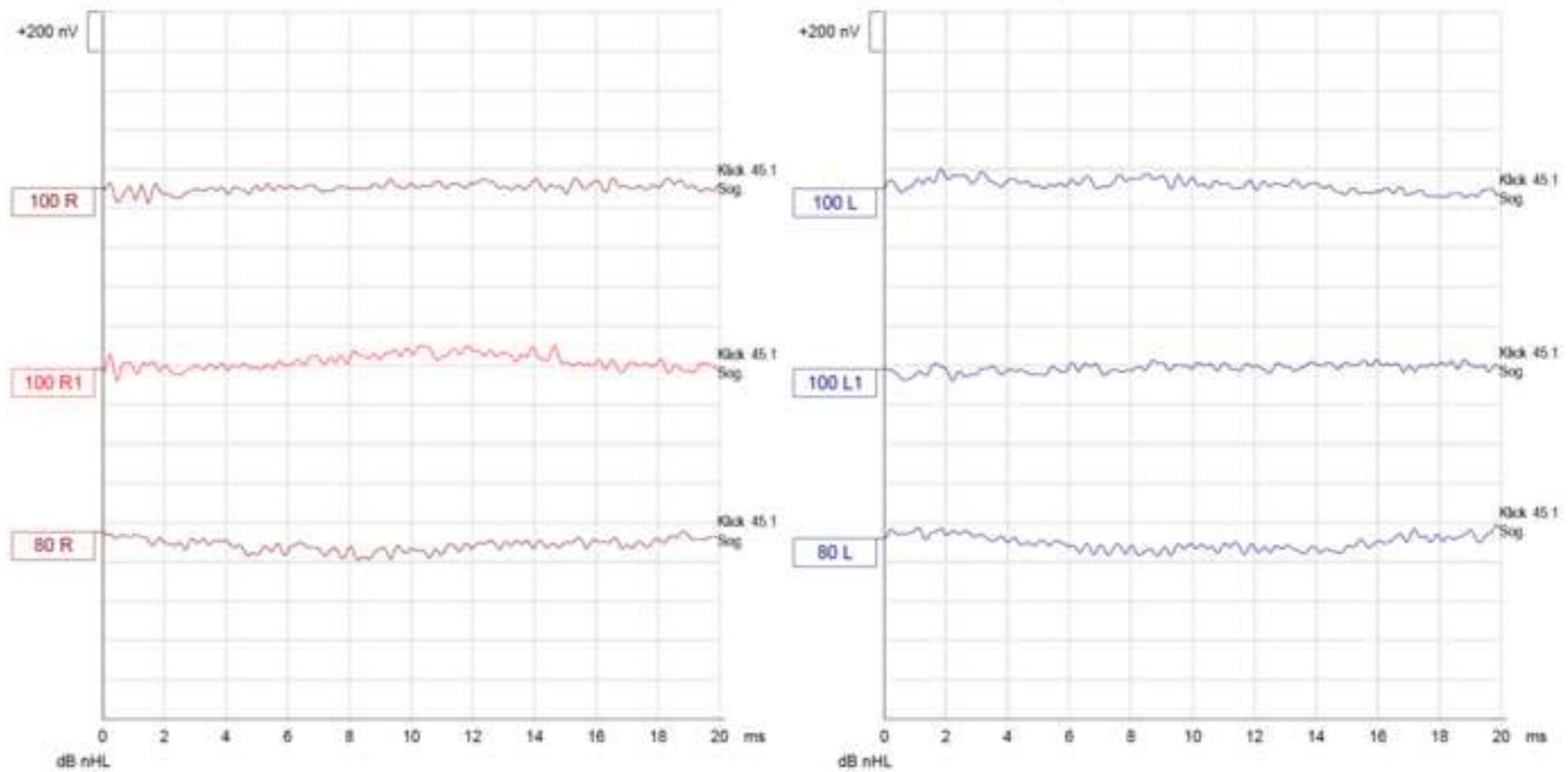
ACKNOWLEDGEMENTS:

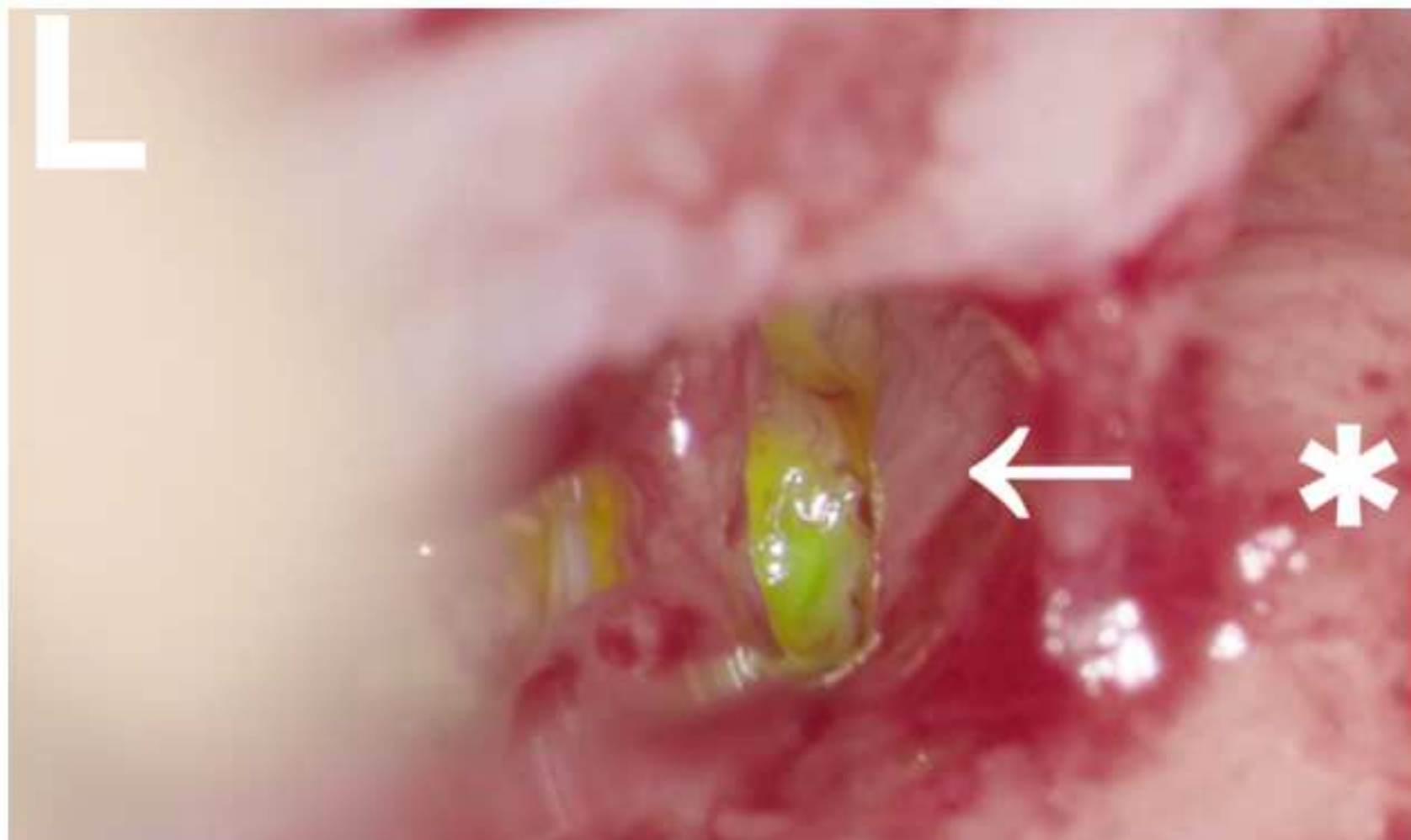
No funding was received.

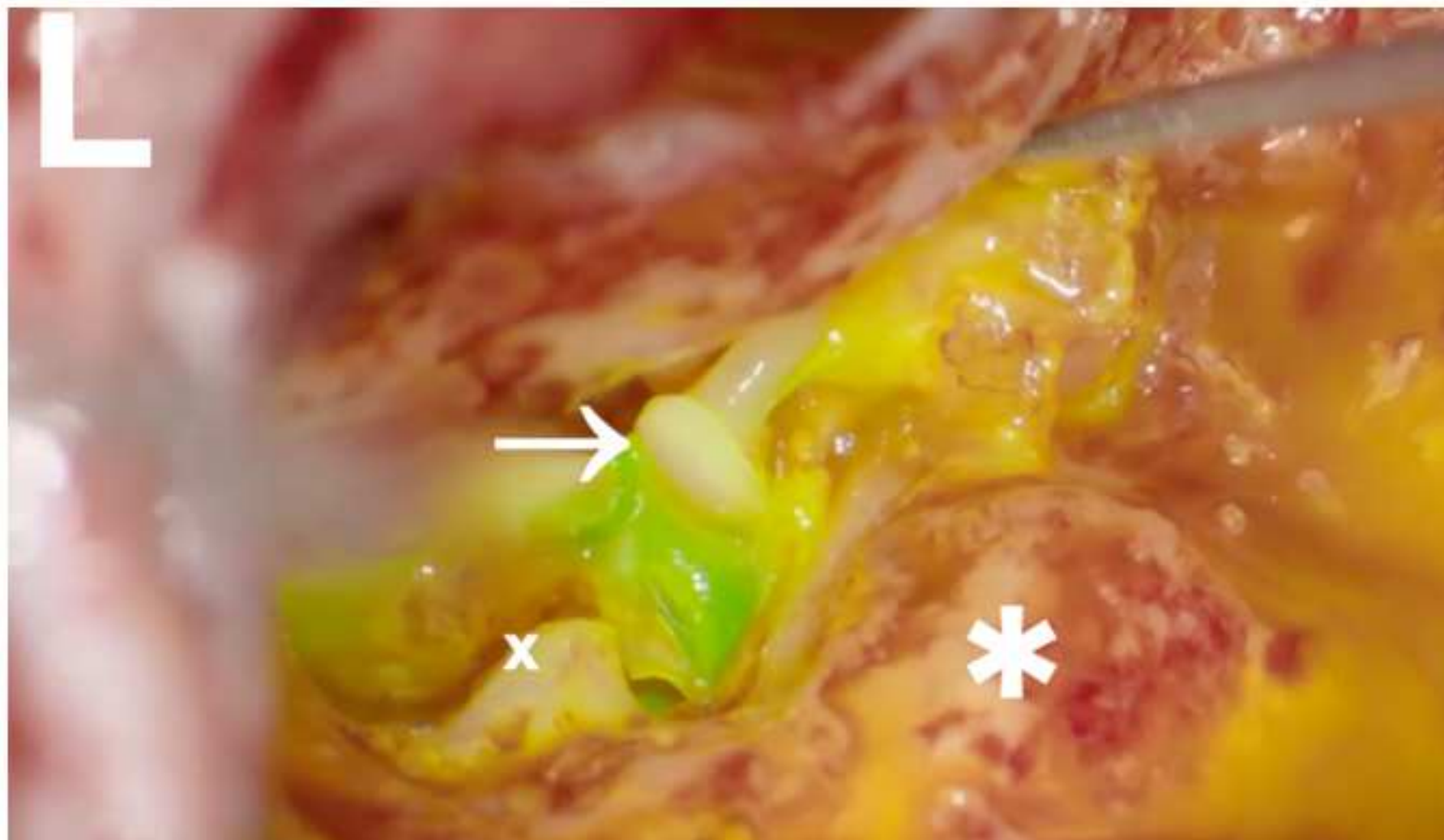
REFERENCES:

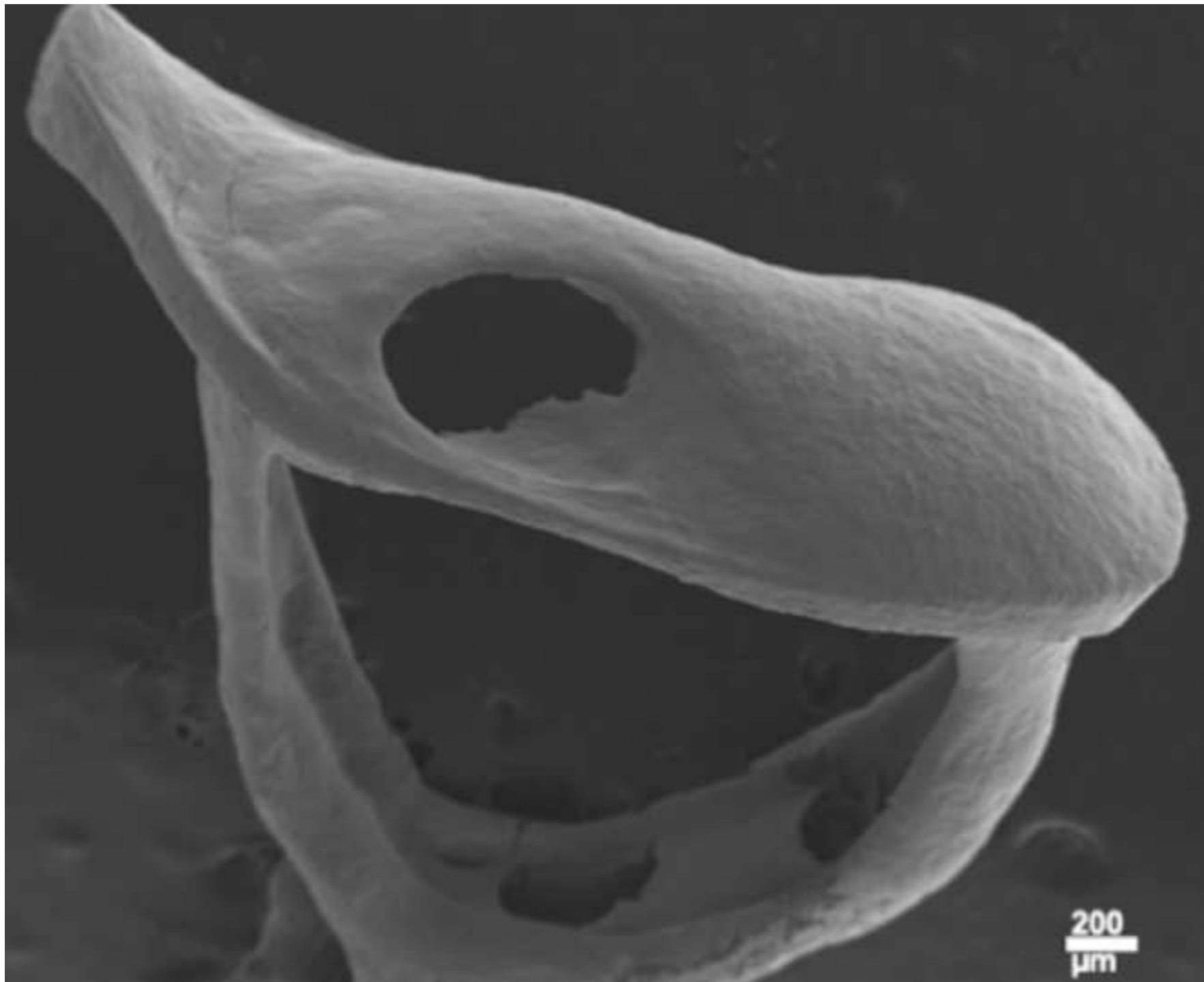
1. Gacek, R. R., Gacek, M. R., Tart, R. Adult spontaneous cerebrospinal fluid otorrhea: diagnosis and management. *The American Journal of Otolaryngology*. **20** (6), 770–776 (1999).
2. Schraven, S. P., Bisdas, S., Wagner, W. Synchronous spontaneous cerebrospinal fluid leaks in the nose and ear. *The Journal of Laryngology and Otology*. **126** (11), 1186–1188 (2012).
3. Raza, S. M. et al. Sensitivity and specificity of intrathecal fluorescein and white light excitation for detecting intraoperative cerebrospinal fluid leak in endoscopic skull base surgery: a prospective study. *Journal of Neurosurgery*. **124** (3), 621–626 (2016).

4. Enghard, A. S., Volgger, V., Leunig, A., Messmer, C. S., Ledderose, G. J. Spontaneous nasal cerebrospinal fluid leaks: management of 24 patients over 11 years. *European Archives of Oto-rhino-laryngology*. **275** (10), 2487–2494 (2018).
5. Sieskiewicz, A., Lyson, T., Rogowski, M., Mariak, Z. Endoscopic management of cerebrospinal fluid rhinorrhea. *The Polish Otolaryngology*. **63** (4), 343–347 (2009).
6. Liu, H. et al. The use of topical intranasal fluorescein in endoscopic endonasal repair of cerebrospinal fluid rhinorrhea. *Surgical Neurology*. **72** (4), 341–5 (2009).
7. Javadi, S. A. H., Samimi, H., Naderi, F., Shirani, M. The use of low- dose intrathecal fluorescein in endoscopic repair of cerebrospinal fluid rhinorrhea. *Archives of Iranian medicine*. **16** (5), 264–266 (2013).
8. Cheng, E., Grande, D., Leonetti, J. Management of spontaneous temporal bone cerebrospinal fluid leak: A 30-year experience. *American Journal of Otolaryngology*. **40** (1), 97–100 (2019).
9. Tandon, S., Singh, S., Sharma, S., Lahiri, A. K. Use of Intrathecal Fluorescein in Recurrent Meningitis after Cochlear Implantation. *Iranian Journal of Otorhinolaryngology*. **28** (86), 221–226 (2016).
10. Warnecke, A. et al. Diagnostic relevance of beta2-transferrin for the detection of cerebrospinal fluid fistulas. *Archives of Otolaryngology - Head & Neck Surgery*. **130** (10), 1178–1184 (2004).
11. Keerl, R., Weber, R. K., Draf, W., Wienke, A., Schaefer, S. D. Use of sodium fluorescein solution for detection of cerebrospinal fluid fistulas: an analysis of 420 administrations and reported complications in Europe and the United States. *The Laryngoscope*. **114** (2), 266–272 (2004).
12. Keerl, R., Weber, R. K., Draf, W., Radziwill, R., Wienke, A. Complications of lumbar administration of 5% sodium fluorescein solution for detection of cerebrospinal fluid fistula. *Laryngo-rhino-otologie*. **82** (12), 833–838 (2003).
13. Harley, B. et al. Endoscopic transnasal repair of two cases of spontaneous cerebrospinal fluid fistula in the foramen rotundum. *Journal of Clinical Neuroscience: Official Journal of the Neurosurgical Society of Australia*. **59**, 350–352 (2019).
14. Wolf, G., Greistorfer, K., Stammberger, H. Endoscopic detection of cerebrospinal fluid fistulas with a fluorescence technique. Report of experiences with over 925 cases. *Laryngo-rhino-otologie*. **76** (10), 588–594 (1997).









Name
Fluorescein ALCON 10% injection solution
External lumbar drain catheter Silverline
Otologic instruments (round knives, hooks, curette, sickle knife, microscissors and microforceps)
Surgical microscope ARRISCOPE 1.0
Consumables <ul style="list-style-type: none">• Water to rinse• Antifog solution• Cotton pads• Cottonoid pledges• Gown• Gloves• Mask• suction tubes• Blade

Company
NOVARTIS
Spiegelberg
Spiggle und Theis
ARRI Medicals

Catalog Number
1467007
ELD33.010.02
NA

Comments
No further specification; instruments can vary between the clinics and their use depends on the surgeon's preferation
No further specification; material of the standard equipment of the operating clinic can be used

JoVE submission JoVE60795 - [EMID:5ae9d09d728ef889]

"Intrathecal application of a fluorescent dye for the identification of cerebrospinal fluid leaks in cochlear malformation"

Rebuttal letter: Response to the reviewer's comments

Dear Reviewers, dear Editors,

thank you very much indeed for your kind time considering our manuscript and video. The resulting changes due to your comments added very much to the value of this work.

Please find enclosed the attention we payed to each of your comments.

Kindest regards,

The authors

Editorial comments from December, 19th :

Comment#1: Please use the **attached word document 60795_R1.docx** to make all edits requested previously (all comments are available in text). Some of the comments were addressed but some were not. Please maintain the format and track all changes. Please do not use the old author-submitted version of your manuscript to make the requested edits.

Response to comment#1: As requested, the corresponding changes were tracked in the attached word document. We hope, that we could address your demands with this version.

Editorial comments from December, 17th :

Comment#1: @01:28 "...about the off-label use of fluorescein. In a first step..."; Separate the words "...fluorescein" and "In a first step..." by about a 1 second. These two phrases need to stand apart so the viewers can reflect briefly on what's been said before moving into the steps of the protocol.

Response to comment#1: The respective change was made to the video

Comment#2: @03:40-05:08 – merge the content from this portion with the Surgery section of the protocol; use the present tense and imperative voice.

Response to comment#2: The respective change was made to the video. The section was merged with the surgical part of the protocol.

Comment#3: @05:10-06:21 Please use the past tense to describe the presented case in the results section.

Response to comment#3: The respective change was made to the video voiceover.

Comment#4: @06:24 The video clip of Dr. Weiss 'pops' in beneath the fading out Conclusion title. Apply a fade-in to the Dr. Nora clip so that it transitions smoothly.

Response to comment#4: The respective change was made to the video.

Comment#5: There is 10 seconds of black at the end of the video. We only need maximum of 3.

Response to comment#5: The respective change was made to the video. We shortened the black part at the end of the video.

Editorial comments from October 29th:

Abstracts: Please remove the subheadings from "Introduction", "Methods", "Results", etc.

Answer to comment: The subheadings were deleted.

Introduction: Please expand your Introduction to include the following: The advantages over alternative techniques with applicable references to previous studies; Description of the context of the technique in the wider body of literature; Information that can help readers to determine if the method is appropriate for their application.

Answer to comment: To our best knowledge, the strength of this protocol is that it is the first description of a clear visualization of CSF leaks of the temporal bone. Our literature review revealed only one case report concerning this issue. The introduction was expanded on this concern.

Protocol Language: The JoVE protocol should be almost entirely composed of numbered short steps (2-3 related actions each) written in the imperative voice/tense (as if you are telling someone how to do the technique, i.e. "Do this", "Measure that" etc.). Any text that cannot be written in the imperative tense may be added as a brief "Note" at the end of the step (please limit notes). Please re-write your ENTIRE protocol section accordingly. Descriptive sections of the protocol can be moved to Representative Results or Discussion. The JoVE protocol should be a set of instructions rather a report of a study. Any reporting should be moved into the representative results

Answer to comment: The protocol was changed using short description steps and the descriptive sections were moved to the results section.

Protocol Detail:

1) Please include an ethics statement before your numbered protocol steps indicating that the protocol follows the guidelines of your institutions human research ethics committee.

Answer to comment 1: The ethics statement was added. Written informed consent of both parents was obtained.

2) Please include specifics of all surgical tools used.

Answer to comment 2: Since Otosurgery is highly variant and varies between different surgical options, we do not determine every single surgical tool since the focus of this video is based on the visualization of the CSF leak. Surgeons considering to adapt the method are aware of the correct surgical access to the mastoid even though the surgical instruments and sealant materials may vary between the clinics.

3) Add pre-surgery preparation steps about patient inclusion/exclusion, sterile draping, anesthesia, muscle relaxation etc.

Answer to comment 3: The according changes were made to the Protocol section.

Protocol Numbering: Please adjust the numbering of your protocol section to follow JoVE's instructions for authors, 1. should be followed by 1.1. and then 1.1.1. if necessary and all steps should be lined up at the left margin with no indentations. Please add a one-line space after each protocol step.

Answer to comment: The corrections were made according to your request.

Discussion: JoVE articles are focused on the methods and the protocol, thus the discussion should be similarly focused. Please ensure that the discussion covers the following in detail and in paragraph form (3-6 paragraphs): 1) modifications and troubleshooting, 2) limitations of the technique, 3) significance with respect to existing methods, 4) future applications and 5) critical steps within the protocol.

Answer to comment: The discussion section was adapted to this concern.

Figures: On Fig 3 it is unclear why each panel has 2 scale bars. Please check.

Answer to comment: The figure was uploaded in an updated version.

Figure/Table Legends: Please expand the legends to adequately describe the figures/tables. Each figure or table must have an accompanying legend including a short title, followed by a short description of each panel and/or a general description.

Answer to comment: The figure legends were adapted.

References:

- 1) Use superscripted numbers for citations.
- 2) At least 10 references must be present.
- 3) Please make sure that your references comply with JoVE instructions for authors. Citation formatting should appear as follows: (For 6 authors or less list all authors. For more than 6 authors, list only the first author then *et al.*): [Lastname, F.I., LastName, F.I., LastName, F.I. Article Title. *Source*. **Volume** (Issue), FirstPage – LastPage, doi:DOI (YEAR).]
- 4) Please spell out journal names.

Answer to comment 1-4: The requested changes to the references were made.

Table of Materials: Please revise the table of the essential supplies, reagents, and equipment. The table should include the name, company, and catalog number of all relevant materials/software in separate columns in an xls/xlsx file. Please include items such as surgical tools, microscope, drugs etc.

Answer to comment: The table was corrected accordingly. No specifications for the surgical tools are given since their use depends on the surgeon's preference. The same applies to the individual anesthesia that has to be chosen according to the patient's history with respect to the anesthesiologist's decision.

Editorial Comments on the Video:

- JoVE's videos are typically step by step protocols that detail a novel procedure that can be repeated, cited, and improved upon. The details are too sparse, the results are muddled into the procedure, and we need some representative results that are separate from the on going commentary.
- Please ensure homogeneity between the written text and the video.
- Please add a distinct protocol and results section. These should be indicated with title cards. Currently the sections are unclear with the results blended in which does not follow our format and style.
- The protocol in the video must be presented in the imperative voice as much as possible with specific case discussions moved into the results section.
- This protocol needs an ethics certification as it involves human subjects. Please add an ethics statement prior to showing the protocol steps.
- Audio is cut off at the beginning at 01:03 ("presented case"). Please fix.
- The English language voice over and commentary "voice" switches tenses. In some cases, the present tense is used; in others, the past tense and past perfect tense. **The voice over should be in present tense consistently.**
- 2:49: Please remove the commercial reference to ArriScope.

Answer to comments: The video was rearranged on these concerns and the complete voiceover was changed.

Comments from Peer-Reviewers:

Reviewer #1:

Manuscript Summary:

This is a good case report with a message to detect and treat CSF leaks in cochlear malformation patients

Major Concerns:

Comment#1: HRCT temporal bone findings prior to surgery are not mentioned?

Answer to comment#1: High resolution computed tomography (HRCT) was performed preoperatively during the diagnostics but did not reveal sincere fracture lines or other hints to reveal the origin of the CSF leak. In the video, we comment on the HRCT findings and the manuscript was adapted respectively.

Comment#2: Which ear is being implanted?

Answer to comment#2: In the presented case the left ear was affected. We clarified this in both, the video and the manuscript

Comment#3: Were you anticipating csf leak preoperatively?

Answer to comment#3: CSF leak was suspected due to the clear secretion from the ear. It was diagnosed in the aural smear with the proof of increased values of beta-transferrin.

Comment#4: Hearing test findings before implant?

Answer to comment#4: Brainstem evoked audiometry was performed and confirmed bilateral deafness. The results are visualized in the video. A corresponding information and figure were added to the manuscript additionally.

Comment#5: MRI findings of patient?

Answer to comment#5: MRI revealed a bilateral temporal bone malformation with aplasia of the right cochlea and a dilatation of the left cochlea and vestibule with absent interscalar septum identical to an incomplete partition type one. This information was added to the written manuscript.

Comment#6: What surgical approach was used to seal the leak site?

Answer to comment#6: In this case, transmastoidal otobasal exploration was indicated. A retroauricular transmastoidal approach was chosen. Mastoidectomy and posterior tympanotomy were performed to access the middle ear space. The leak was easily identified at the stapes footplate. The content of the manuscript was clarified on this concern.

Minor Concerns:

Comment#7: What material was used to seal the leak site?

Answer to comment#7: Fascia from the temporal muscle, temporal muscle and absorbable collagen fibrin sealant patches are used to seal the CSF leak. We added this information to the manuscript.

Comment#8: Any manoeuvre done after injecting dye?

Answer to comment#8: In this particular case, no special manoeuvre was necessary since good visualization was easily achieved. In the literature, sometimes Trendelenburg positioning is recommended for cases where the intracranial pressure needs to be increased. From our experience, this method may be waived.

Comment#9: Allergic testing done with fluorescein?

Answer to comment#9: Since allergic reactions and side effects are reported to be rare and surgery is performed under controlled conditions in the operation theatre and since the dye was applied immediately prior surgery, no testing was done. An according section was added to the discussion.

Reviewer #2:

Manuscript Summary:

Nice article with preliminary data on the use of fluorescein to assess for the localization of lateral skull base leaks.

Minor Concerns:

Comment#1: Perhaps discuss a few minor concerns you need to address with intrathecal fluorescein use (e.g., seizure risk).

Answer to comment#1:

The application of fluorescein was described as a safe method in skull base surgery using endoscopic techniques. Side effects are assumed to depend on the dose ¹⁰. Though the use is “off-label” no side-effects were observed in all of our patients receiving intrathecal fluorescein and the dye can be applied repeatedly. No severe long term side effects are to be expected ¹¹. We added a comment to this in the discussion section of the manuscript.

Comment#2: Also, what type of microscope is used? I use an LED or Xenon bulb. What type of light do I need to use if I borrow your technique?

Answer to comment#2: The use of both, Xenon and LED bulbs is possible. For intraoperative visualization of the fluorescent dye they are covered with a special filter that selects the emission light. In the case of fluorescein sodium, a blue light filter needs to be applied. The emitted fluorescence (green in the case of fluorescein) is again filtered from the remaining colors by the use of a spectral emission filter. The filter systems are available from the respective companies.