A Pseudo-Outbreak of *Pseudomonas putida* and *Stenotrophomonas maltophilia* in a Bronchoscopy Unit

Maribel Botana-Rial^a Virginia Leiro-Fernández^a Manuel Núñez-Delgado^a
Maximiliano Álvarez-Fernández^b Susana Otero-Fernández^b
Helena Bello-Rodríguez^c Carlos Vilariño-Pombo^a Alberto Fernández-Villar^a

^aBronchopleural Unit, Department of Pneumology, and Departments of ^bMicrobiology and ^cEpidemiology, Hospital Álvaro Cunqueiro, NeumoVigo I+i Research Group, Research Institute Galicia Sur (IIS Galicia Sur) EOXI Vigo, Vigo, Spain

Key Words

Bronchoscope · Pseudo-outbreak · Automated endoscope reprocessor · *Pseudomonas putida* · *Stenotrophomonas maltophilia*

Abstract

Background: Endoscopes represent the medical devices most commonly linked to health care-associated outbreaks and pseudo-outbreaks. Most of the recent outbreaks and pseudo-outbreaks have resulted from contaminated automated endoscope reprocessors (AER) or the use of damaged or malfunctioning bronchoscopes or contaminated equipment. **Objectives:** We investigated a pseudo-outbreak of *Pseudomonas putida* and *Stenotrophomonas maltophilia* recovered from bronchial washing (BW) specimens obtained during bronchoscopy in a bronchoscopy unit. **Methods:** Samples were obtained from environmental surfaces in the endoscopy suite, bronchoscopes, and bronchoscopic dispensable material, and specimens of cleaning solutions, cleaning brushes, the AER, and the ultrasound system were sent for bacterial culture. Medical records were reviewed to identify possible infections after a bronchoscopy. **Results:** *P. putida* and *S. maltophilia* were isolated from BW samples of

39 patients. The bronchoscopy models Olympus BF-1T160 and BF-160 were contaminated. Both bronchoscopes and other contaminated material (cleaning brushes, diluted cleaning solutions, and the sink) were isolated, but new cases continued to appear. The AER was recently installed, and new connections were used for the water lines and new tubes were connected to the AER. Initially, specimens were obtained from the external circuits and the internal walls of the AER. Finally, cultures were made from the filters on the water lines, and growth of *P. putida* and *S. maltophilia* was found. The investigation revealed that the BW specimens were contaminated because sterile saline was injected by means of the biopsy port of the bronchoscope and was recovered through the same channel by means of the proximal suction port. No patients developed clinical signs or symptoms of infection, but the positive cultures did lead to treatment of 21 patients. **Conclusions:** We described a pseudo-outbreak related to a contaminated bronchoscope because of inadequate installation of the AER for used new water lines and because the new tubes were connected to the AER. The antibacterial filters of the AER used tap water, and this may have contained low levels of microorganisms. No serious clinical complications derived from this pseudo-outbreak.

© 2016 S. Karger AG, Basel

Introduction

Flexible bronchoscopy is one of the most common and useful procedures performed by chest clinicians. It is a widely used technique for the diagnosis and therapy of pulmonary diseases [1, 2]. Although the risk of endoscopy-related transmission of infection is estimated to be very low, more health care-associated infections are related to contaminated endoscopes than to any other medical device. However, most of the recent outbreaks and pseudo-outbreaks have resulted from contaminated automated endoscope reprocessors (AER) or the use of damaged or malfunctioning bronchoscopes or contaminated equipment, including rinsing tanks, tubing, antibacterial filters on water lines, and cleaning brushes [3–5]. Most reports refer to pseudo-outbreaks as the isolation of organisms in bronchoscopy specimens due to colonization or contamination of the bronchoscope rather than true patient-to-patient transmission ('true infection') [5, 6]. The most common organisms implicated in bronchoscopy-related pseudo-outbreaks include bacterial pathogens such as *Pseudomonas aeruginosa* or *Serratia marcescens*, contagious and noncontagious mycobacteria, and environmental fungi. *Pseudomonas*, *Legionella*, and *Serratia* species and *Mycobacterium tuberculosis* are among the most common organisms reported in true infections [5, 7]. Therefore, standardizing guidelines on cleaning and high-level disinfection or sterilization is important. In general, endoscope reprocessing includes pre-cleaning (at the bedside), leak testing, manual cleaning, high-level disinfection, rinsing, drying, and storage. An AER could be used to perform leak testing and high-level disinfection and to rinse flexible endoscopes [7].

In this report, we describe a pseudo-outbreak of *Pseudomonas putida* and *Stenotrophomonas maltophilia* in an endoscopy unit related to contamination of the suction port of a bronchoscope as the result of inadequate installation of the AER.

Materials and Methods

At the time of the investigation, the Complejo Hospitalario Universitario de Vigo (CHUVI) was a 1,000-bed tertiary care hospital in Spain. Approximately 1,200 bronchoscopy procedures were performed annually, for both diagnostic and therapeutic purposes. The endoscopy department had two bronchoscopy suites which had been inaugurated the previous month. An epidemiologic investigation was conducted to determine the cause of *P. putida* and *S. maltophilia* growth in bronchial washing (BW) specimens of patients who underwent flexible bronchoscopy between February 7 and March 4, 2013. In February, the microbiology lab-

oratory reported the growth of *P. putida* and *S. maltophilia* from separate BW specimens obtained on different days. The endoscopy unit cleans bronchoscopes according to the manufacturers' recommendations and national guidelines [1, 7]. The cleaning and disinfection processes, as well as bronchoscope maintenance and repair records, were evaluated. All bronchoscopes in the endoscopy unit were inspected for evidence of loose ports or others defects, and culture samples were obtained by instilling sterile saline into the distal (biopsy) port and collecting samples in both an antegrade and a retrograde manner from the same biopsy channel. Samples were obtained from environmental surfaces in the endoscopy suite and from dispensable bronchoscopic material (valves and connections), and specimens of cleaning solutions, cleaning brushes, the AER (model EDT3 Olympus; only of the external circuits and of the internal walls), and the ultrasound system were sent for bacterial culture. Bacterial species were identified by an automated system. After species identification, all available isolates were submitted to molecular typing with REP-PCR BOX-A. Medical records of patients whose bronchoscopy specimens yielded *P. putida* or *S. maltophilia* were reviewed and the following variables were recorded: the date of bronchoscopy, results of bronchoscopic specimen culture for routine respiratory pathogens, the bronchoscopy device used, and symptoms or signs and other microbiological and radiological studies to identify respiratory tract colonization or infections possible in the following 14 days after a bronchoscopy and whether antibiotic therapy was initiated. The local ethics committee approved this study.

Statistical Analysis

The global results are reported as percentages and absolute frequencies for the qualitative variables and as the mean value and standard deviation (SD) for the quantitative variables. The data were analyzed with the use of the program Statistical Package for the Social Sciences version 14.0 (SPSS, Chicago, Ill., USA).

Results

During the study period, 154 bronchoscopies were performed with 8 different bronchoscope models. The bronchoscopies were performed in accordance with the standard procedure, monitoring the patient in a supine position and using conscious sedation (midazolam $\pm$ fentanyl). The nasal route was used in most cases, and 2% lidocaine was employed as an anesthetic. Of 39 cases with positive BW specimens, 28 (71.8%) were positive for *P. putida* and *S. maltophilia*, 9 (23.1%) only for *P. putida*, and 2 (5.1%) only for *S. maltophilia*. Fifteen patients had mixed cultures for other microorganisms (table 1). The patients' median age was 63 years (range 51–75), and 28 (71.8%) were male. Eleven patients (28.2%) were immunosuppressed: 7 patients had lymphoma, 1 patient had human immunodeficiency virus infection, 2 patients were treated with immunosuppressive therapy, and 1 patient received chemotherapy for lung cancer.

Table 1. Results of cultures of specimens from environmental sources, bronchoscopes, and BW

Source and type of specimen	<i>P. putida</i> or <i>S. maltophilia</i> isolated on culture
Cleaning brushes	Yes
Diluted cleaning solutions	Yes
Sink	Yes
AER	No
Ultrasound system bronchoscope	Yes
Bronchoscopes UC-180 and BF-PE2	No
Bronchoscopes BF-1T160 and BF-160	Yes
Other cultures positive in BW	Patients (n = 15), n
<i>Pneumocystis jirovecii</i>	2
<i>Aspergillus</i> species, influenza virus, and <i>Mycobacterium lentiflavum</i>	2
Influenza virus	1
Methicillin-resistant <i>Staphylococcus aureus</i>	1
<i>Mycobacterium lentiflavum</i>	5
<i>Proteus</i> species	1
<i>Candida</i> species, <i>Escherichia coli</i>	1
<i>Candida</i> species, <i>Mycobacterium lentiflavum</i>	1
<i>Mycobacterium tuberculosis</i>	1

A targeted investigation was initiated at the bronchoscopy unit. Environmental cultures were also taken. Initially, in different specimens from the environmental sources, growth of *P. putida* and/or *S. maltophilia* was observed (table 1). The positive isolates were associated with procedures performed with two Olympus bronchoscopy models in particular: the BF-1T160 model in 12 (30.8%) and the BF160 model in 18 (46.2%) of the cases. Only one procedure (2.6%) was associated with the Olympus bronchoscopy model BF-1T180. The bronchoscopes and the contaminated material were isolated. The contaminated bronchoscopes were isolated and replaced with other ones of the same model. Though we adhered to new preventive recommendations, new cases continued to appear with the models BF-1T160 and BF-160.

The AER was examined again. It was recently installed, and new connections were used for the water lines and new tubes were connected to the AER. The antibacterial filters of the AER used tap water, and this may have contained low levels of microorganisms. Initially, specimens were obtained from the external circuits and the internal walls of the AER. Finally, cultures were made from the filters on the water lines; a grossly visible biofilm was

found, as well as growth of *P. putida* and *S. maltophilia*, in the water tank of the AER (fig. 1). We developed a hypothesis to explain these findings: the investigation revealed that the BW specimens were contaminated because sterile saline was injected by means of the biopsy port and was recovered through the same channel by means of the suction port. The filters on the water lines were cleaned and disinfected. During follow-up, samples were obtained from bronchoscopes, and the isolates were negative.

Molecular typing was performed on all patients isolated. Repetitive PCR typing showed that the isolates recovered from the patients were indistinguishable. The two organisms were related with the same strain (fig. 2). No serious clinical complications derived from this pseudo-outbreak. The patients developed neither clinical signs or symptoms of infection nor radiological alterations, but 21 patients (53.8%) were treated with antibiotics (*P. putida* and *S. maltophilia* were sensitive to quinolones) and 13 (33.3%) were receiving antibiotic treatment before bronchoscopy.

Discussion

Although infectious complications of flexible bronchoscopy are uncommon, outbreaks and pseudo-outbreaks related to bronchoscopy have been reported [3–5, 8–13]. The proper method for high-level disinfection or sterilization of bronchoscopes is well described in current guidelines [1, 2, 7]. However, the use of an AER is not specifically mentioned in the various guidelines or recommendations. An AER improves standardization of the reprocessing steps, reduces personnel exposure to high-level disinfectants and infectious material, and lowers the possibility of human errors.

Kenters et al. [5] present an overview of publications about outbreaks and pseudo-outbreaks related to contamination of flexible endoscopes. There are four publications related to failure due to a defective or malfunctioning AER [14–17]. Furthermore, the function of the water filtration system in the AER might not be reliable in providing bacteria-free water. Because tap water may contain low levels of microorganisms, some authors have suggested that only sterile water (which may be prohibitively expensive) or filtered water using an AER be used [5, 6]. The suggestion that only sterile water or filtered water should be used is not consistent with the published guidelines, which allow tap water with an alcohol rinse and forced-air drying [7].

Fig. 1. **a, b** Samples collected from the external circuits, the internal walls and filters on the water lines, or the tank of the AER. **c, d** Biofilms with *P. putida* and *S. maltophilia* growth found in the water tank of the AER.

Fig. 2. Results of pulsed-field gel electrophoresis demonstrating the clonal pattern of the isolates from 6 patients (2–5, 7, and 8) in the pseudo-outbreak.

Stigt et al. [18] describe a pseudo-outbreak of *S. maltophilia* caused by failure of the disinfectant to sterilize ultrasound scopes. Recently, one article described pseudo-outbreaks of *S. maltophilia* isolated in water used to flush bronchoscopes after decontamination, and another arti-

cle related pseudo-outbreaks of *Mycobacterium gordonae* isolated in tap water and in the water supply channel of a washer-disinfector [19, 20]. Prolonged storage in humid conditions seems to enhance the growth of microorganisms.

We described a pseudo-outbreak related to only two models of bronchoscopes, BF-1T160 and BF-160. Only one procedure was associated with the Olympus bronchoscopy model BF-1T180. The authors believe that the Olympus bronchoscopy model BF-1T180 was mistakenly registered, because the same model was used during the study period and was not associated with positive isolates. In addition, the other bronchoscopes in use during the study period were not associated with positive cultures.

One hypothesis is that probably the design of the bronchoscopes, in the proximal part of the suction channel, favors the condition of dampness suitable for the growth of these bacteria. Persistent bacterial contamination of the biopsy port was probably not a result of a breach of the cleaning and disinfection guidelines, because the hospital's reprocessing procedures were determined to be satisfactory. Drying the endoscope is essential for greatly reducing the chance of recontamination of the endoscope by microorganisms that may be present in the rinse water.

A pseudo-outbreak is defined as isolation of transmitted organisms in bronchoscopy specimens obtained from patients without evidence of a specific infection. The specimens most commonly isolated are Gram-negative bacteria, *Mycobacterium* or *Legionella* species [5, 7]. We did not find clear evidence that any patients started developing respiratory tract infections after having undergone bronchoscopy with the contaminated bronchoscopes; thus, we considered this a pseudo-outbreak. It is remarkable that 11 patients in this series were immunosuppressed. However, a possible limitation to this study is that positive cultures led to treatment directed at *P. putida* or *S. maltophilia* in 21 patients who likely did not need antibiotic therapy. We emphasized the fact that the BW specimens were contaminated because sterile saline

was injected by means of the biopsy port and was recovered through the same channel by means of the suction port. We were able to identify this pseudo-outbreak before more clinical infections occurred in patients.

In conclusion, outbreaks and pseudo-outbreaks associated with bronchoscopes continue to occur. Meticulous attention to proper cleaning and disinfection of endoscopes is critical for their prevention. Early detection of contamination would be easier if standardized periodic microbiological testing were included in the guidelines. Because AER are often used for flexible endoscope reprocessing, they should be included in the guidelines. Periodic maintenance of bronchoscopes and AER as well as new installations should always be carried out as the manufacturer advises.

References

- Du Rand IA, Blaikley J, Booton R, Chaudhuri N, Gupta V, Khalid S, Mandal S, Martin J, Mills J, Navani N, Rahman NM, Wrightson JM, Munawar M; British Thoracic Society Bronchoscopy Guideline Group: British Thoracic Society guideline for diagnostic flexible bronchoscopy in adults: accredited by NICE. *Thorax* 2013;68(suppl 1):i1–i44.
- Casal RF, Ost DE, Eapen GA: Flexible bronchoscopy. *Clin Chest Med* 2013;34:341–352.
- Peaper DR, Havill NL, Aniskiewicz M, Callan D, Pop O, Towle D, Boyce JM: Pseudo-outbreak of *Actinomyces graevenitzi* associated with bronchoscopy. *J Clin Microbiol* 2015;53:113–117.
- Cosgrove SE, Ristaino P, Caston-Gaa A, Fellerman DP, Nowakowski EF, Carroll KC, Orens JB, Perl MT, Maragakis LL: Caveat emptor: the role of suboptimal bronchoscope repair practices by a third-party vendor in a pseudo-outbreak of *Pseudomonas* in bronchoalveolar lavage specimens. *Infect Control Hosp Epidemiol* 2012;33:224–229.
- Kenters N, Huijskens EGW, Meier C, Voss A: Infectious diseases linked to cross-contamination of flexible endoscopes. *Endosc Int Open* 2015;3:E259–E265.
- Weber DJ, Rutala WA: Lessons learned from outbreaks and pseudo-outbreaks associated with bronchoscopy. *Infect Control Hosp Epidemiol* 2012;33:230–234.
- Metha AC, Prakash UBS, Garland R, Haponik E, Moses L, Schaffner W, Silvestri G: American College of Chest Physicians and American Association for Bronchology consensus statement: prevention of flexible bronchoscopy-associated infection. *Chest* 2005;128:1742–1755.
- Kirschke DL, Jones TF, Craig AS, Chu PS, Mayernick GG, Patel JA, Schaffner W: *Pseudomonas aeruginosa* and *Serratia marcescens* contamination associated with a manufacturing defect in bronchoscopes. *N Engl J Med* 2003;348:214–220.
- Ece G, Erac B, Limoncu MH, Baysak A, Oz AT, Ceylan KC: *Stenotrophomonas maltophilia* pseudo-outbreak at a university hospital bronchoscopy unit in Turkey. *West Indian Med J* 2014;63:59–61.
- Silva CV, Magalhães VD, Pereira CR, Kawagoe JY, Ikura C, Ganc AJ: Pseudo-outbreak of *Pseudomonas aeruginosa* and *Serratia marcescens* related to bronchoscopes. *Infect Control Hosp Epidemiol* 2003;24:195–197.
- Schuetz AN, Hughes RL, Howard RM, Williams TC, Nolte FS, Jackson D, Ribner BS: Pseudo-outbreak of *Legionella pneumophila* serogroup 8 infection associated with a contaminated ice machine in a bronchoscopy suite. *Infect Control Hosp Epidemiol* 2009;30:461–466.
- Srinivasan A, Wolfenden LL, Song X, Mackie K, Hartsell TL, Jones HD, Diette GB, Orens JB, Yung RC, Ross TL, Merz W, Scheel PJ, Haponik EF, Perl TM: An outbreak of *Pseudomonas aeruginosa* infections associated with flexible bronchoscopes. *N Engl J Med* 2003;348:221–227.
- Rosengarten D, Block C, Hidalgo-Grass C, Temper V, Gross I, Budin-Mizrachi A, Berkman N, Benenson S: Cluster of pseudoinfections with *Burkholderia cepacia* associated with a contaminated washer-disinfector in a bronchoscopy unit. *Infect Control Hosp Epidemiol* 2010;31:769–771.
- Chroneou A, Zimmerman SK, Cook S, Willey S, Eyre-Kelly J, Zias N, Shapiro DS, Beamis JF Jr, Craven DE: Molecular typing of *Mycobacterium chelonae* isolates from a pseudo-outbreak involving an automated bronchoscope washer. *Infect Control Hosp Epidemiol* 2008;29:1088–1090.
- Sorin M, Segal-Maurer S, Mariano N, Urban C, Combet A, Rahal JJ: Nosocomial transmission of imipenem-resistant *Pseudomonas aeruginosa* following bronchoscopy associated with improper connection to the Steris System 1 processor. *Infect Control Hosp Epidemiol* 2001;22:409–413.
- Vanhems P, Gayet-Ageron A, Ponchon T, Bernet C, Chayvialle JA, Chemorin C, Morand L, Bibollet MA, Chevallier P, Ritter J, Fabry J: Follow-up and management of patients exposed to a flawed automated endoscope washer-disinfector in a digestive diseases unit. *Infect Control Hosp Epidemiol* 2006;27:89–92.
- Mean M, Mallaret MR, Bichard P, Shum J, Zarski JP: Gastrointestinal endoscopes cleaned without detergent substance following an automated endoscope washer/disinfector dysfunction. *Gastroenterol Clin Biol* 2006;30:665–668.
- Stigt JA, Wolfhagen MJ, Smulders P, Lammers V: The identification of *Stenotrophomonas maltophilia* contamination in ultrasound endoscopes and reproduction of decontamination failure by deliberate soiling tests. *Respiration* 2015;89:565–571.
- Waite TD, Georgiou A, Abrishami M, Beck CR: Pseudo-outbreaks of *Stenotrophomonas maltophilia* on an intensive care unit in England. *J Hosp Infect* 2016;92:392–396.
- Scorzolini L, Mengoni F, Mastroianni CM, Baldan R, Cirillo DM, De Giusti M, Marinelli L, Cottarelli A, Fattorini L, Vullo V: Pseudo-outbreak of *Mycobacterium gordonae* in a teaching hospital: importance of strictly following decontamination procedures and emerging issues concerning sterilization. *New Microbiol* 2016;39:25–34.