Disinfection of the flexible fibreoptic bronchoscope against *Mycobacterium tuberculosis* and *M. gordonae*

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**ABSTRACT** In view of recent reports of contamination of the fibreoptic bronchoscope by tuberculous and non-tuberculous mycobacteria, we evaluated the disinfecting properties of aqueous glutaraldehyde (Cidex) and an iodophor (Prepodyne) against *Mycobacterium tuberculosis* and *M. gordonae*. We found that a 15 minute disinfection procedure with either agent, coupled with initial vigorous mechanical cleaning of the bronchoscope and its accessories, is a quick and reliable method for preventing the contamination of the bronchoscope with mycobacteria.

Although flexible fibreoptic bronchoscopy has become accepted as a safe diagnostic and therapeutic procedure, one of its complications is infection. Recently there have been reports of pulmonary tuberculosis caused by a contaminated bronchoscope. There are also reports of contamination by non-tuberculous mycobacteria in specimens obtained through the flexible fibreoptic bronchoscope, which may lead to an erroneous diagnosis and inappropriate treatment. Leers reviewed the procedures for disinfecting flexible fibreoptic bronchoscopes, but there is no consensus about the most reliable and rapid method for disinfecting the fibreoptic bronchoscope against mycobacterial organisms. We therefore set out to determine whether glutaraldehyde and iodophor, when used in the manner recommended by one fibreoptic bronchoscope manufacturer (Olympus), will disinfect a substantial inoculum of *Mycobacterium tuberculosis* and *M. gordonae*.

**METHODS**

The bronchoscope (Model B1T, Olympus) was contaminated on 12 different occasions: six times with *M. gordonae* and six times with *M. tuberculosis*. The fibreoptic bronchoscope and accessories were disinfected with either aqueous glutaraldehyde or iodophor and each disinfectant was tested against each organism on three different occasions.

**PREPARATION OF THE INOCULUM**

Stock cultures of *M. gordonae* (Trudeau Mycobacterium Culture (TMC) No 1318 strain W911) and *M. tuberculosis* (TMC No 201 and strain H37RA) were grown to turbidity in Dubos medium and were diluted in 100 ml of sterile 7% albumin solution. *M. gordonae* is a common contaminant which is non-pathogenic and safe to handle and H37RA is an attenuated strain of *M. tuberculosis*. The 7% albumin solution was used to simulate the viscous character of bronchial secretions. With the use of a 0.001 calibrated semiquantitative loop, a 7H-11 culture plate was implanted with the inoculating solutions and incubated at 35°C in 7% carbon dioxide until growth appeared. The colonies were counted and the number was multiplied by the dilution factor (×1000). The concentration of organisms was 4.2 × 10^9 *M. gordonae/ml and 3.12 × 10^9 *M. tuberculosis/ml.

**SIMULATING CONTAMINATION**

The following pieces of equipment were used in the experiment: (1) the automatic aspiration adaptor, located at the proximal end of the channel sleeve and used to control suction during bronchoscopy; (2) the cleaning brush, used for cleaning the suction channel; (3) the manual suction adaptor, which replaces the automatic aspiration adaptor during cleaning to irrigate the channel; and (4) the distal tip of the fibreoptic bronchoscope and its suction channel.

To contaminate the fibreoptic bronchoscope (which had been gas sterilised with ethylene oxide at twice atmospheric pressure for six hours and
allowed to aerate for at least 36 hours), the tip was immersed in 100 ml of inoculating solution. The solution was aspirated through the channel into the automatic aspiration adaptor with a syringe and allowed to stand for 15 minutes. The aspirated solution was then discarded, and material for culture was obtained from the various components of the fibreoptic bronchoscope to serve as a positive control. The inoculating process was then repeated. To simulate the clinical setting, in which time may elapse between using and cleaning the fibreoptic bronchoscope, the suction channel was allowed to remain undisturbed for 15 minutes.

DISINFECTION
The bronchoscope, the automatic aspiration adaptor, and the cleaning brush were all cleaned in a manner similar to that recommended by the manufacturer: the fibreoptic bronchoscope was immersed in sterile water, the suction channel was cleaned by passing the cleaning brush through the bronchoscope five times, and the channel was vigorously irrigated with about 10 ml of sterile water. The automatic aspiration adaptor was disassembled (fig), immersed in sterile water, and mechanically cleaned with the cleaning brush. The cleaning brush was rubbed vigorously between gloved fingers to remove all debris.

The solutions whose ability to kill mycobacteria were studied were (1) a commercially available undiluted iodine solution containing 1% titratable iodine (Prepodyne, AMSCO Medical Products Division, Erie, Pennsylvania), and (2) freshly activated 2% alkaline glutaraldehyde (pH 8-8.5, Cidex, Surgikos Inc, Arlington, Texas). The cleaning brush, the distal 40 cm of the fibreoptic bronchoscope, and the automatic aspiration adaptor were immersed in either glutaraldehyde or iodophor. After 15 minutes of contact with the disinfectant solution all equipment was thoroughly rinsed in sterile water and material was obtained for culture from the fibreoptic bronchoscope, automatic aspiration adaptor, and cleaning brush.

CULTURE TECHNIQUE
Material was obtained at the following times: (1) before contamination; (2) after immersion in inoculating solution, to demonstrate that contamination had taken place; and (3) after disinfection.

The distal 5 cm of the fibreoptic bronchoscope was immersed in a 35 ml glass incubation tube containing 20 ml of sterile Dubos medium. The medium was aspirated with a syringe through the suction channel and the manual suction adaptor, allowed five minutes of contact time, and then returned to the original glass tube. The automatic aspiration adaptor was disassembled and placed in a specimen cup containing 20 ml of sterile Dubos medium for five minutes. The medium was then transferred to a sterile 35 ml glass incubation tube. The tip of the cleaning brush was immersed in 20 ml of Dubos medium contained in a 35 ml glass tube for five minutes.

Cultures were incubated in 7% carbon dioxide at 35°C for five weeks. Growth was detected by making smears of turbid broths or by centrifuging clear broths at 3000 g for 15 minutes in 50 ml plastic centrifugation cups and making smears of the sediment. The smears were stained with Auramine O - Rhodamine B acid fast stain and examined by fluorescent microscopy. The demonstration of fluorescent bacilli in culture material was interpreted as a "positive" culture and their absence was interpreted as a "negative" culture.

All work was performed under a laminar airflow hood with sterile gloves, masks, and gowns.

Results
The culture results showed that each component of the equipment (bronchoscope, automatic aspiration adaptor and cleaning brush) was on each occasion contaminated after exposure to the inoculating material. After disinfection with either glutaraldehyde or iodophor, however, the cultures for either *M tuberculosis* or *M gordonae* on all pieces of equipment were on each occasion negative. Thus, mechanical cleaning of the fibreoptic bronchoscope...
and automatic aspiration adaptor with the cleaning brush and complete contact with either disinfectant solution for 15 minutes eliminated all mycobacteria.

Discussion

Controversy still exists about which method of rapid, cold disinfection is most reliable in preventing the transmission of mycobacteria by the fiberoptic bronchoscope. Garcia de Cabo and colleagues used benzalkonium chloride followed by alcohol to disinfect the bronchoscope against several pathogenic organisms, including M tuberculosis. Reports show, however, that benzalkonium chloride fails to kill M tuberculosis17-19 and alcohols are unreliable disinfectants against tubercle bacilli in sputum.20 Suratt and coworkers used a more complex protocol of hexachlorophene, povidone-iodine (Betadine), ethanol, and water. Recent studies, however, have shown that Betadine or hexachlorophene alone may be inactive against M tuberculosis.

Our study indicates that a simple three step procedure of mechanical cleaning followed by 15 minutes' disinfection with either aqueous glutaraldehyde or iodophor (Prepodyne) and rinsing with sterile water effectively eliminated M tuberculosis and M gordonae from an artificially contaminated fiberoptic bronchoscope. We tried to approximate the clinical conditions under which the bronchoscope is contaminated. Highly concentrated inocula of M tuberculosis and M gordonae were mixed with albumin to simulate bronchial secretions contaminated by these organisms. Since there may be a short delay in practice before disinfecting the bronchoscope, we waited for 15 minutes before starting decontamination. Our results may, however, apply only to disinfection performed shortly after using the bronchoscope, since it is possible that prolonged drying might interfere with cleaning and disinfection. The H37RA strain of M tuberculosis used in this study is attenuated, but we have no reason to suspect that the disinfection procedure would be any less effective against more virulent mycobacteria.

All parts of the fiberoptic bronchoscope should be cleaned mechanically before chemical disinfection and particular attention must be given to the components of the automatic aspiration adaptor (fig), which may retain contaminated material.

The disinfectants most recently recommended by the Olympus Company are an iodophor (Wescodyne) or glutaraldehyde (Cidex, Glutarex, or Sonacide). The chemical agents and periods of disinfection used in the present study appear to be quite satisfactory. Aqueous alkaline glutaraldehyde is highly mycobactericidal and does not harm the fiberoptic bronchoscope.25 It has a slight odour, and since it may be irritating to the eyes and skin it should be rinsed carefully from the instrument and hands. Iodophors are non-irritating but may stain.26 Careful removal by wiping the bronchoscope, particularly the distal viewing tip, with a gauze pad and sterile water can prevent discoloration. Recent information suggests that not all iodophors are equally active against mycobacteria,11 so it seems reasonable, until more information is available, to use one which is known to have mycobactericidal activity. Finally the water used for rinsing the bronchoscope should be sterile, to avoid inadvertent contamination with organisms such as M gordonae, which may be present in tap water.

Pappas et al recently reported the apparent failure of glutaraldehyde to disinfect the fiberoptic bronchoscope of M chelonei and Pseudomonas aeruginosa. Two bronchoscopes were found to be damaged and they considered the source of the original contamination was tap water or glutaraldehyde solutions, which allowed colonisation of the puncture sites in the suction channel of the bronchoscope. Tap water is a well known source of contamination and it is likely that infectious organisms collected in these damaged sites, out of reach of the cleaning brush and disinfectant. M chelonei has been isolated from low (0-2%) concentration glutaraldehyde packing solutions; but this does not argue against the use of a much higher concentration (2%) as a disinfecting agent, which in the present study gave satisfactory results.

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References