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Major Article

Residual moisture and waterborne pathogens inside flexible endoscopes: Evidence from a multisite study of endoscope drying effectiveness

Cori L. Ofstead MSPH *, Otis L. Heymann BA, Mariah R. Quick MPH, John E. Eiland RN, MS, Harry P. Wetzler MD, MSPH

Ofstead & Associates, Inc, Saint Paul, MN

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Background: Endoscopy-associated infection transmission is frequently linked to inadequate reprocessing. Residual organic material and moisture may foster biofilm development inside endoscopes. This study evaluated the effectiveness of endoscope drying and storage methods and assessed associations between retained moisture and contamination.
Methods: Endoscope reprocessing, drying, and storage practices were assessed at 3 hospitals. Researchers performed visual examinations and tests to detect fluid and contamination on patient-ready endoscopes.
Results: Fluid was detected in 22 of 45 (49%) endoscopes. Prevalence of moisture varied significantly by site (5%; 83%; 85%; $P < .001$). High adenosine triphosphate levels were found in 22% of endoscopes, and microbial growth was detected in 71% of endoscopes. *Stenotrophomonas maltophilia, Citrobacter freundii*, and *Lecanicillium lecanii/Verticillium dahliae* were found. Retained fluid was associated with significantly higher adenosine triphosphate levels ($P < .01$). Reprocessing and drying practices conformed with guidelines at 1 site and were substandard at 2 sites. Damaged endoscopes were in use at all sites.
Conclusions: Inadequate reprocessing and insufficient drying contributed to retained fluid and contamination found during this multisite study. More effective methods of endoscope reprocessing, drying, and maintenance are needed to prevent the retention of fluid, organic material, and bioburden that could cause patient illness or injury.
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Endoscopy-associated pathogen transmission has been recognized as a patient safety issue.[1-6] Outbreaks have been linked to contaminated duodenoscopes,[7-9] gastroscopes,[10,11] cystoscopes,[12-14] ureteroscopes,[15,16] and bronchoscopes.[4,17-21] Although current guidelines permit the reuse of flexible endoscopes after cleaning and high-level disinfection (HLD), contamination often remains after reprocessing, and repeated reprocessing has proven inadequate to eliminate bioburden.[22-28]

Residual fluid may be retained inside endoscopes after HLD.[10,26,27,29,30] This is problematic because wet environments facilitate growth of gram-negative bacteria[31] and other potential pathogens.[4,32,33] Previous investigations found that fully-reprocessed endoscopes harbored waterborne pathogens,[4,22,31-34] including *Pseudomonas aeruginosa*[10,12,19,21] and *Stenotrophomonas maltophilia*.[19,35,36] Reprocessing guidelines describe drying as critically important,[37-39] but there is no consensus among experts and guideline-issuing bodies on best practices for endoscope drying and storage.[31,37-39] Alfa and Sitter reported that 10 minutes of purging with forced air reduced gram-negative bacilli in endoscope channels.[40] However, this method has not been widely embraced because it requires space, equipment, and time. Instead, many institutions rely on alcohol flushes and brief air purges before hanging endoscopes vertically in hopes that residual fluid will drain out or evaporate. This study evaluated the effectiveness of endoscope drying and storage methods used in the field and assessed associations among drying methods, retained moisture, and residual contamination levels.

* Address correspondence to Cori L. Ofstead, MSPH, Ofstead & Associates, Inc, 1360 Energy Park Drive, Suite 300, Saint Paul, MN 55108-5300.
E-mail address: cori@ofsteadinsights.com (C.L. Ofstead).
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MATERIALS AND METHODS

Design

This prospective, multisite, observational study was conducted in 2017 in 3 multispecialty hospitals in the United States that were members of large healthcare systems and accredited by The Joint Commission. No human subjects were involved. The institutional review board (IRB) of each site determined that the study was a quality assurance project, and thus no IRB review was needed. Data were collected during site visits by the research team.

Sampling methods

When researchers arrived, site personnel selected fully-reprocessed endoscopes for study inclusion that were representative of their inventory and had been stored for >24 hours. Sterile swabs moistened with sterile, deionized water were used to sample ports and distal ends before placement into transport media (480/482C ESwabs; COPAN Diagnostics Inc., Murrieta, California). Channel samples were obtained using the flush-brush-flush technique[26,31,41–43] with sterile, deionized water and a sterile channel swab (Healthmark Industries, Fraser, Michigan). Channel effluent and swabs were transported in Dey-Engley neutralizing broth (Hardy Diagnostics, Santa Maria, California). Adenosine triphosphate (ATP) tests were performed on effluent, biopsy ports, control handles, and distal ends (CleanTrace ATP Water and ATP Surface; 3M Company, St. Paul, Minnesota). Researchers maintained strict aseptic technique while sampling.

Samples were transported to Food and Drug Administration-registered, International Organization for Standardization-certified microbiology laboratories. Samples were extracted and concentrated using 0.45-µm filters that were placed onto tryptic soy and blood agar plates for 5-7 days' incubation at 28-32°C. Colony-forming units (CFUs) were counted daily. Final counts represented the total per channel or surface site. Species identification was pursued when gram-negative bacteria or mold were found. To confirm the validity of sampling and testing methods, positive and negative control tests were performed on samples from clinically used, pre-cleaned gastrointestinal endoscopes and sterile water, swabs, and neutralizing broth.

Moisture tests and visual examinations

After sampling, endoscopes were re-reprocessed by site personnel in accordance with institutional protocols and stored for 24-48 hours before visual examinations. Retained fluid and other irregularities were photographed with a camera and borescopes designed to examine channels and ports (0.8-mm Ultra-Thin HQ Micro Borescope; Medit Inc., Winnipeg, Canada; 2.3-mm and 3.2-mm Flexible Inspection Scope; Healthmark Industries). Whether or not fluid was observed, moisture tests were performed after the borescope examination using a sterile swab and chemical indicator for water (Hydrion Humidicator Paper; Micro Essential Laboratory Inc., Brooklyn, New York). A pilot test confirmed the indicator's ability to detect 10 µL of water transferred by micropipette onto swabs.

Assessment of reprocessing practices

At each site, researchers observed endoscope reprocessing, drying, and storage practices. To objectively assess storage cabinet cleanliness, ATP tests were conducted on the door handles, interior walls, and floors of cabinets.

Statistical analysis

Excel 2016 (Microsoft Corporation, Santa Rosa, California) and SPSS Statistics version 21 (IBM Corporation, Armonk, New York) software were used for data analysis. Differences in moisture prevalence and microbial growth were assessed using Fisher's exact test. ATP differences between sites and by endoscope type were examined with the Kruskal-Wallis test. Post-hoc site differences were evaluated with the Mann-Whitney test as were ATP differences by presence or absence of retained moisture. The expected ATP level was <40 relative light units (RLUs) for materials free of residual contamination, and a validated benchmark of 200 RLU was used to distinguish high ATP levels in endoscopes.[42,44] To reduce the occurrence of type 1 errors, the threshold for statistical significance was set at .01.

RESULTS

Researchers examined 45 patient-ready endoscopes (43 Olympus; 2 Karl Storz; Table 1). The study involved 13 colonoscopes, 12 gastroscopes, 5 duodenoscopes, 3 cystoscopes, 3 ureteroscopes, 3 endoscopic ultrasound endoscopes, 3 bronchoscopes, 2 intubation endoscopes, and 1 endobronchial ultrasound bronchoscope.

Retained fluid

Droplets were observed inside 21 of 45 channels (47%) (Fig 1), with significant differences by site (A: 10 of 12 [83%]; B: 0 of 20 [0%]; C: 11 of 13 [85%]; $P < .001$). Test strips detected water in 22 of 45 (49%) endoscopes, with significant differences by site (A: 10 of 12 [83%]; B: 1 of 20 [5%]; C: 11 of 13 [85%]; $P < .001$; Table 1). The positive predictive value of test strips was 95.5% (95% confidence interval [CI]: 77.2%–99.9%); the negative predictive value was 100% (95% CI: 85.2%–100%). Oily fluid was observed on several endoscopes, and the fluid subsequently tested negative for water (Fig 2). Whitish-blue, powdery residue and dried white "water spots" were commonly observed on external surfaces.

ATP tests

Ten of 45 (22%) endoscopes had ATP levels >200 RLU (A: 3 of 12 [25%]; B: 1 of 20 [5%]; C: 6 of 13 [46%]; $P = .012$; Table 1). ATP levels for 31 of 45 (69%) endoscopes were ≥40 RLU (A: 7 of 12 [58%]; B: 12 of 20 [60%]; C: 12 of 13 [92%]; $P = .087$; Table 1). Differences between Sites B and C were significant for the maximum ATP values per endoscope ($P = .003$) and surface ATP results ($P = .002$). Retained moisture was associated with higher maximum ATP levels ($P < .01$).

Microbial cultures

Microbial growth was detected in 32 of 45 (71%) endoscopes (A: 11 of 12 [92%]; B: 10 of 20 [50%]; C: 11 of 13 [85%]; $P = .30$; Table 1). At Site B, microbial growth was found in 10 of 16 (62%) high-level disinfected endoscopes and in 0 of 4 (0%) sterilized endoscopes (B-17, B-18, B-19, B-20). Colony counts were ≥10 CFU for 4 of 12 (33%) endoscopes at Site A and for 6 of 13 (46%) endoscopes at Site C. Colonies were too numerous to count for 2 of 12 (17%) endoscopes at Site A and for 5 of 13 (38%) endoscopes at Site C. Microbes at those sites included *Lecanicillium lecanii/Verticillium dahliae* (mold) and *S. maltophilia* (Fig 3). Although colony counts were lower at Site B (all <10 CFU), waterborne bacteria and other potential pathogens (e.g., *S. maltophilia* and *Citrobacter freundii*) were found. Endoscopes with retained moisture were more likely to have microbial growth, but differences were not significant ($P = .028$). No

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