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Major Article

How to better monitor and clean irregular surfaces in operating rooms: Insights gained by using both ATP luminescence and RODAC assays

Olivia Ellis MS, MPH, M(ASCP)[CM], PHM, REHS [a], Hilary Godwin PhD [a,b], Maren David MS [c], Dan J. Morse MS [c], Romney Humphries PhD, ABMM, M(ASCP)[CM] [d,e], Daniel Z. Uslan MD, MBA, FIDSA, FSHEA [a,f,*]

[a] *UCLA Fielding School of Public Health, Department of Environmental Health Sciences, Los Angeles, CA*
[b] *Institute of the Environment and Sustainability, University of California, Los Angeles, CA*
[c] *3M Infection Prevention, St. Paul, MN*
[d] *Department of Pathology & Laboratory Medicine, David Geffen School of Medicine, University of California, Los Angeles, CA*
[e] *Accelerate Diagnostics, Tucson, AZ*
[f] *Division of Infectious Disease, Department of Medicine, David Geffen School of Medicine, University of California, Los Angeles, CA*

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Background: A major limitation to developing evidence-based approaches to infection prevention is the paucity of real-time, quantitative methods for monitoring the cleanliness of environmental surfaces in clinical settings. One solution that has been proposed is adenosine triphosphate (ATP) bioluminescence assays, but this method does not provide information about the source of the ATP.
Materials/Methods: To address this gap, we conducted a study in which ATP bioluminescence was coupled with traditional RODAC sampling and matrix-assisted laser desorption/ionization-time-of-flight mass spectrometry to assess which organisms were viable and present. Using this mixed assessment approach, we evaluated cleaning of 5 different types of high-touch surfaces (overhead lights, door handles, anesthesia keyboards, mattresses, and side tables) in operating rooms.
Results: Whether surfaces tested cleaner after turnaround than they did before turnaround depended on the surface type. Before and after cleaning, flat, covered surfaces (mattresses and side tables) were more likely to pass as "clean" by ATP assay than uncovered, irregularly shaped surfaces (overhead lights, door handles, and anesthesia keyboards). Irregularly shaped surfaces were more likely to pass by RODAC assay than by ATP assay after cleaning.
Conclusion: Our results suggest that irregularly shaped surfaces in operating rooms may require enhanced covering, cleaning, and monitoring protocols compared to more regularly shaped surfaces.
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BACKGROUND

Cleaning and disinfection of environmental surfaces and patient care equipment are essential components of infection prevention in the healthcare setting.[1] The environment serves as a reservoir for healthcare-associated infections (HAIs).[2] Furthermore, well-documented evidence links the transmission of pathogens to contaminated hospital surfaces.[3,4] Therefore, frequent and effective disinfection of "high-touch surfaces" is a critical step in ensuring patient safety.[5]

Evidence-based recommendations for cleaning, disinfection, and sterilization in hospitals are published by the Healthcare Infection Control Practices Advisory Committee (HICPAC) at the Centers for Disease Control and Prevention.[1] As defined by HICPAC, disinfection is the process by which many or all pathogenic microorganisms, other than spores, are eliminated from inanimate objects.[1] This process begins with appropriate environmental cleaning and removal of organic or inorganic debris from surfaces with the use of an Environmental Protection Agency-registered hospital-grade disinfectant.[1] Hospital operating rooms (ORs) have policies in place that require environmental services staff and perioperative staff to

* Address correspondence to Daniel Z. Uslan, MD, MBA, FIDSA, FSHEA, David Geffen School of Medicine, University of California, Los Angeles, CA 90095-1772.
E-mail address: duslan@mednet.ucla.edu (D.Z. Uslan).
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follow cleaning guidelines established by HICPAC and the Association of peri-Operative Registered Nurses, in an effort to reduce HAIs.[1,4] ORs must be disinfected between each procedure, and every OR should be terminally disinfected at the end of each day.[4]

Despite these disinfection practices, approximately 1 of every 25 hospitalized patients develops an HAI.[6] Of these, surgical site infections (SSIs) account for 31% of all HAIs and are associated with the greatest additional healthcare cost, estimated at $2.5-$10 billion annually in the United States.[2,6,7] ORs contain a variety of high-touch surfaces that can serve as fomite reservoirs of microorganisms and pathogens. Inadequate cleaning of these environmental surfaces likely contributes to SSIs.[2,4,8] Therefore, methodologies are needed that would allow for routine environmental monitoring for microbial contamination and potential pathogens present on a broad range of different surfaces types in an OR setting.

Previous studies have shown that adenosine triphosphate (ATP) bioluminescence monitoring has the potential to provide rapid quantitative measures of effective cleaning in hospitals.[9,10] Environmental ATP monitoring offers an advantage over microbiologic methods, which use an array of swabs and sponges accompanied by various broths and agars and are time and labor intensive.[11] ATP assays have an advantage over other methods (e.g., fluorescent markers) for studying the thoroughness of cleaning and disinfection processes in that ATP assays can be used to monitor contamination in real time and are semi-quantitative.[12] Although ATP is an excellent biomarker, ATP detection can be caused by either viable organisms or non-viable cellular material and does not differentiate between pathogenic and non-pathogenic organisms.[11] As a result, the magnitude of the signal obtained from this technique is not a direct measure of pathogenicity or patient risk.[11] Still, ATP monitoring is regularly performed in several industries, including aerospace, food and beverage, ecology, cosmetics, and clinical.[11] The food safety industry and the planetary protection division of the National Aeronautics and Space Administration have reported that the ATP assay is the best currently available, real-time option for monitoring cleanliness, since contamination is not random and organisms can survive on surfaces for months.[11,13-16]

In this study, we used ATP monitoring, in combination with a replicate organism detection and counting (RODAC) assay and matrix-assisted laser desorption/ionization-time-of-flight (MALDI-TOF) mass spectrometry, to assess cleaning effectiveness of irregularly shaped surfaces compared to regularly shaped surfaces in a clinical environment. Specifically, we compared results for regularly shaped (flat), covered surfaces (mattresses and side tables) and for irregularly shaped, uncovered surfaces (overhead lights, door handles, and anesthesia keyboards) to determine how surface types affected the utility of ATP bioluminescence testing.

MATERIALS AND METHODS

We compared the results obtained from real-time ATP monitoring of 5 different types of high-touch surfaces in operating rooms after cleaning to high-touch surfaces before cleaning in the same room. The surfaces were also assessed after cleaning using a traditional quantitative microbiologic method (RODAC plates) in addition to the ATP assay. MALDI-TOF mass spectrometry was used to identify organisms isolated on the RODAC plates from samples taken from surfaces and the air.

Sampling strategy

Samples were collected in ORs in a 520-bed teaching hospital over 5 consecutive weeks on 24 separate occasions. Sampling was performed before and after turnaround cleaning between procedures. Some ORs were sampled on more than 1 separate occasion. Hospital environmental services staff performed regular turnaround cleaning alongside perioperative anesthesia staff according to existing hospital protocols and were aware that sampling was taking place. Turnaround cleaning was always performed between patients, using either quaternary ammonia with microfiber cloths and/or disposable bleach disinfectant wipes. The disinfectant was left to dry on each surface before sampling. Before turnaround cleaning, ATP sampling was performed on the 5 different high-touch OR surfaces (door handles, overhead lights, anesthesia keyboards, side tables, and patient mattresses) in each room. After turnaround cleaning, ATP and bacterial load (RODAC) sampling was performed on the same high-touch OR surfaces in each of the same rooms. To ensure rigor in sampling methodologies, samples were collected by a trained public health microbiologist, not by the individuals actually responsible for cleaning the rooms. Additionally, RODAC air samples were collected after turnaround cleaning near the door of each OR.

Surface sampling

ORs were sampled for viable bacterial contamination within 20 minutes of the completion of surgeries and turnaround cleaning procedures. RODAC plates were used to culture bacteria from surfaces and contained 5 neutralizers, (sodium bisulfate, sodium thioglycollate, sodium thiosulfate, lecithin, and polysorbate 80), which inactivate disinfectant agents. Our ATP assay did not contain these neutralizers. To sample, the lid of a RODAC plate was removed, and the agar surface was pressed to the test surface for 10 seconds. In cases where the surface was curved, the plate was gently rolled such that the entire agar surface met the test surface over a 10-second period. The lids of the RODAC plates were snapped back into place, inverted, and incubated aerobically at 35°C ± 2°C for 48 ± 4 hours. Visible colonies were enumerated, and the number of colony-forming units (CFUs)/plate was recorded. Cleaning compliance was assessed using previously established cutoff benchmarks of 64 CFU/plate: pass (≤2.5 CFU/cm^2) and fail (>2.5 CFU/cm^2).[17-20] Three colonies from each plate were selected and subcultured, to individual blood agar plates (BAPs; Hardy Diagnostics), for isolation following in-house standard operating procedures and identified with Biomerieux VITEK-2 MALDI-TOF mass spectrometry.

Air sampling

Air samples were collected near the floor, adjacent to the door opening to the hallway corridor, of each OR after cleaning using a MicroBio MB2 portable impactor (flow rate = 100 L/min) loaded with a RODAC plate. Air samples were collected for 5 minutes for a total volume of 0.5 L.[3] Three colonies from each air plate were subcultured to BAP and identified, per in-house standard operating procedures, with Biomerieux VITEK-2 MALDI-TOF mass spectrometry.

ATP sampling

OR surfaces were swabbed for ATP both before and after turnaround cleaning with 3M CleanTrace ATP surface swabs, and relative light units (RLUs) were measured using a 3M Clean-Trace NGi luminometer. All high-touch surfaces were sampled by swabbing a ~ 25-cm^2 area, similar to that of a RODAC plate. RLUs were obtained according to the manufacturer's specifications.[15] Based on previously published studies, the RLU benchmarks used to assess cleaning compliance were: pass ≤ 250 RLU and fail > 250 RLU.[9,15] The percentage of samples that passed or failed for each type of surface is summarized in Table 1.

MALDI-TOF mass spectrometry

Samples were prepared by subculturing individual colonies from the RODAC plates to BAPs. The BAPs were incubated aerobically for

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