Swab Type, Moistening, and Preenrichment for *Staphylococcus aureus* on Environmental Surfaces[▽]

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We compared five swabs, dry or premoistened and with or without preenrichment, to detect surface contamination with *Staphylococcus aureus*. Sensitivities varied based on swab type, as follows: 71.9% and 75% for rayon, 71.2% for cotton, 81.3% for polyester, and 53.2% for calcium alginate. Preenrichment improved sensitivity (80%, versus 61.3% for direct-plated specimens), as did premoistening (83.4%, versus 57.5% for dry swabs). All of the premoistened, preenriched swabs were positive.

It is believed that transmission of *Staphylococcus aureus* in the health care setting occurs through transfer from colonized or infected individuals, transfer from the hands of health care workers, and contact with contaminated objects in the environment (1, 5). *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* (MRSA) have been shown to persist for weeks on many items in the health care setting, including patient care equipment, uniforms, computer keyboards, cellular phones, and identification (ID) badges (6). Monitoring environmental contamination has been recommended as a measure to direct cleaning and decontamination efforts in order to control outbreaks and reduce transmission (2, 4).

Environmental surveillance is typically performed on surfaces by use of a sterile swab to recover organisms from surfaces. Commercially available swabs are made of cotton, rayon, polyester (including Dacron), or calcium alginate material at the end of a plastic, metal, or wooden shaft. Other surface collection methods include the use of wipe sampling devices (such as synthetic fabrics or sponges), quantitative cultures using contact plates, and ATP bioluminescence (3, 9, 10). No standard method for environmental monitoring exists; premoistening of the swab tip with a sterile solution is often reported and preenrichment of samples is inconsistently reported in studies of environmental contamination (7).

In order to compare methods of environmental-sample collection, we compared five swabs in terms of detection of *S. aureus* from environmental surfaces, impact of premoistening the swab tip, and effect of preenrichment.

Eight representative surfaces (computer keyboard, computer mouse, door knob, mobile-phone keypad, faucet handle, light switch, and two flooring samples) were included in the study. Five swabs were used in random sequence: a commercially available rayon-tipped BBL CultureSwab collection and transport swab (catalog no. 220093; BD Diagnostic Systems, Sparks, MD); a cotton-tipped applicator with a wooden shaft (Q-Tip; Kendall Healthcare Products, Mansfield, MA); a plas-

tic-shaft, polyester-tipped applicator (part no. 36816; Solon, Solon, ME); an aluminum-shaft, calcium alginate nasopharyngeal applicator (part no. 36600; Solon, Solon, ME); and a rayon swab used for the recovery of fastidious organisms (Quest Diagnostics, Pittsburgh, PA). Sterile 0.9% sodium chloride (USP; Baxter Healthcare, Deerfield, IL) was used to premoisten each swab when required.

Each surface was disinfected and then contaminated in a biosafety cabinet by spraying a known solution of *S. aureus* (ATCC 25923) in Trypticase soy broth (TSB) (BD, Sparks, MD) at approximately 1.28×10^5 CFU/cm². Surfaces were allowed to air dry for 24 h prior to the surface collection. A 5-by 5-cm template was placed in the sampling area to approximate the desired sampling surface. The first series of sampling was done with swabs as supplied by the manufacturer ("dry swab"), and then sampling was repeated using a swab that had been premoistened in 0.9% normal sterile saline ("wet swab").

In order to replicate handling procedures at our facility, all swabs were held at room temperature for 8 h and then streaked onto mannitol salt agar (MSA) (BD, Sparks, MD). Following inoculation of the first plate, swabs were preenriched by being placed in 5 ml of TSB (BD, Sparks, MD) for 12 h and then replated to MSA. Incubation was performed at 37°C, and plates were examined for growth at 12, 24, and 48 h. Fisher's exact test was used to calculate statistically significant differences by swab type and by preenrichment status.

There were a total of 140 samples available for analysis. Rayon-tipped swabs and polyester-tipped, plain rayon, and cotton-tipped applicators had similar positive rates (71.9%, 71.2%, 81.3%, and 75%, respectively). Calcium alginate nasopharyngeal applicators had the lowest overall recovery (53.2%; $P = 0.02$ versus rayon swab).

Preenrichment markedly improved the rate of detection, with 64/80 (80%) of preenriched samples positive, compared to 49/80 (61.3%) of direct-plated specimens ($P < 0.01$). Despite the additional time required for processing, preenriched swabs were also positive earlier in the test process, and qualitatively, more growth occurred on preenriched specimens.

Premoistening the swab tip also improved recovery, with 67/80 (83.4%) of wet swabs positive and 46/80 (57.5%) of dry swabs positive ($P < 0.01$). Regardless of the swab type, 100% of premoistened, preenriched samples were positive.

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This study highlights the importance of careful technique when performing environmental sampling. Consistent with findings from other authors (7), we found that sensitivity increased with use of preenrichment regardless of the type of swab used for collection. This was observed despite using the same swab for direct-plating and preenrichment steps, which would be expected to reduce bacterial counts in the second step. It is possible that preenrichment allows organisms recovered from dry surfaces to enter a growth phase prior to plating on selective media. While premoistening of the swab tip has not been universally adopted, these findings suggest that this should be routine practice. Future studies should examine the impact of recovery of staphylococcus *in vivo* using premoistened swabs or wetting of mucosal surfaces.

One drawback of this study is that it was conducted in a controlled environment in which a single strain was introduced. Since it is unlikely that surface contamination with a single species occurs in clinical settings, these results should be duplicated in an actual clinical environment. The surfaces in this study were contaminated with a much higher concentration of bacteria than is typically observed in nature, and we expect that these results may overestimate the sensitivity of each method, which makes the variability even more striking. Second, we did not test differences depending on the type of surface. Moistening of the swab tip may be more beneficial on heavily textured or irregular surfaces than on hard, flat surfaces, which could have implications for monitoring contamination on surfaces such as mobile computing platforms or ergonomic keyboards (8, 11). The results may also be influenced by swab pressure, swab angle, duration of contact, and stroke pattern. While we attempted to control for these variables by having a single technician obtain samples in a controlled environment, these factors may also influence swab performance.

In summary, this study demonstrates the importance of specimen collection methods, specifically swab type and premoistening, and the improvement in sensitivity by using preenrich-

ment for the recovery of staphylococci from environmental surfaces.

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REFERENCES

1. Boyce, J. M. 2007. Environmental contamination makes an important contribution to hospital infection. *J. Hosp. Infect.* **65**(Suppl. 2):50–54.
2. Cooper, R. A., C. J. Griffith, R. E. Malik, P. Obee, and N. Looker. 2007. Monitoring the effectiveness of cleaning in four British hospitals. *Am. J. Infect. Control* **35**:338–341.
3. Dancer, S. 2004. How do we assess hospital cleaning? A proposal for microbiological standards for surface hygiene in hospitals. *J. Hosp. Infect.* **56**:10–15.
4. Dancer, S. J. 2008. Importance of the environment in methicillin-resistant *Staphylococcus aureus* acquisition: the case for hospital cleaning. *Lancet Infect. Dis.* **8**:101–113.
5. Harris, A. D. 2008. How important is the environment in the emergence of nosocomial antimicrobial-resistant bacteria? *Clin. Infect. Dis.* **46**:686–688.
6. Kramer, A., I. Schwebke, and G. Kampf. 2006. How long do nosocomial pathogens persist on inanimate surfaces? A systematic review. *BMC Infect. Dis.* **6**:130.
7. Moore, G., and C. Griffith. 2007. Problems associated with traditional hygiene swabbing: the need for in-house standardization. *J. Appl. Microbiol.* **103**:1090–1103.
8. Po, J. L., R. Burke, C. Sulis, and P. C. Carling. 2009. Dangerous cows: an analysis of disinfection cleaning of computer keyboards on wheels. *Am. J. Infect. Control* **37**:778–780.
9. Sherlock, O., N. O'Connell, E. Creamer, and H. Humphreys. 2009. Is it really clean? An evaluation of the efficacy of four methods for determining hospital cleanliness. *J. Hosp. Infect.* **72**:140–146.
10. Weese, J. S. 2007. Environmental surveillance for MRSA, p. 201–208. In Y. Ji (ed.), *Methods in molecular biology: methicillin-resistant Staphylococcus aureus (MRSA) protocols*. Springer, New York, NY.
11. Wilson, A. P. R., P. Ostro, M. Magnussen, and B. Cooper. 2008. Laboratory and in-use assessment of methicillin-resistant *Staphylococcus aureus* contamination of ergonomic computer keyboards for ward use. *Am. J. Infect. Control* **36**:e19–e25.