



ABSTRACT

Effective disinfection of medical surfaces is crucial in preventing healthcare-associated infections. The objective of this study was to compare two techniques for transferring bacteria, specifically *Staphylococcus epidermidis*, from contaminated medical surfaces to agar plates for growth assessment. The first technique involved imprinting the contaminated surface directly onto the agar plate, while the second technique utilized a sterile swab to pick up bacteria and transfer them to the agar plate. Results indicated a higher and more consistent percent of bacterial transfer occurred using the imprint technique compared to the swab technique. Consequently, the imprint technique was selected for further investigation to quantify results related to the disinfection of contaminated medical surfaces. This study underscores the importance of selecting appropriate bacterial transfer techniques for accurate assessment of surface disinfection efficacy in healthcare settings.

MATERIALS

- *Staphylococcus epidermidis*
- Tryptic Soy Agar Plates
- Difco Nutrient Broth #3 (Media)
- 200, 1000 uL Pipettes and Tips
- 5 mL Overnight Tubes
- 1.5 mL Microfuge Tubes (Dilutions)
- Circular Polypropylene Surfaces (d = 1.75", t = 0.06")
- Sterile Swab
- MATLAB CFU Counting Code



Figure 1: Count Plate



Figure 2: Imprint Plate

METHODS

Control

- Perform and plate dilutions from an overnight sample to determine which dilution produces a significant number (250-500) of countable colony forming units (CFUs). The dilution factor that provided this count was 10^{-4} .
- With this dilution, create a count plate which will serve as a comparison to the results of recollection testing.

Swab Recollection Method

- Using the 10^{-4} dilution that was previously determined, place 20 uL of the sample onto a polypropylene surface.
- Soak the sterile swab in the specified volume of sterile media, including volumes of 50, 75, 100, and 150 uL.
- Use the sterile swab to pick up the bacteria from the surface.
- Swab the recollected bacteria onto an agar plate.
- Place the agar plate in an incubator set to 37°C and allow the sample to incubate for 24 hours before recording results.

Imprint Recollection Method

- Using the 10^{-4} dilution that was previously determined, place 20 uL of the sample onto a polypropylene surface.
- Use a sterile spreader to evenly distribute the bacteria across the entire area of the polypropylene surface.
- Imprint surface onto an agar plate and wait 30 seconds.
- Remove the surface from the agar plate and discard in biosafety bag.
- Place the agar plate in an incubator set to 37°C and allow the sample to incubate for 24 hours before recording results.

Counting Methods

- After initially performing colony counts by hand or trying to utilize online programs, our team developed our own computer program using MATLAB's image processing software to accurately count colonies.
- An example of the output of this program can be seen in Figure 3 to the right. One feature that made this program more efficient was the fact that the total CFU count was displayed in the title of output. This addition greatly enhanced our ability to quickly and accurately perform colony counts and may be useful in future applications.

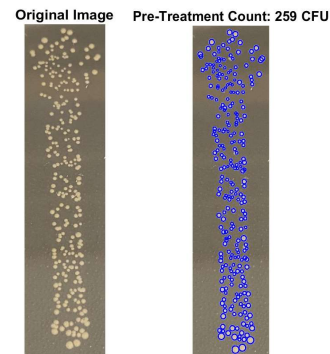
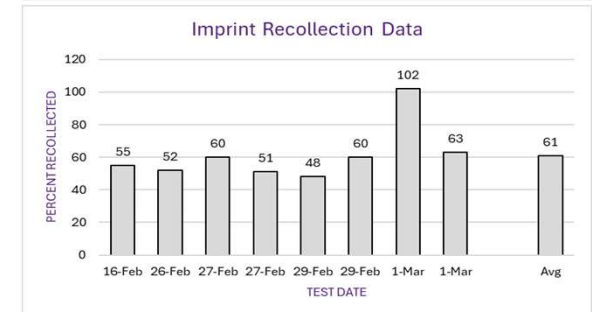
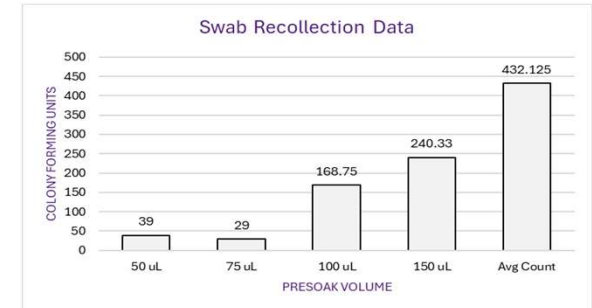


Figure 3: MATLAB Output

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RESULTS



CONCLUSIONS

The results of this research indicate the imprint recollection method is more effective than the pre-soaked swab method. The Swab Recollection Data results indicate that increasing the pre-soaking of the swab led to a higher collection of bacteria. The imprint technique, where bacteria are spread before imprinting, yielded a recollection percentage of approximately 61%. The results of this study corroborate the Campos-Murguía study, which claims the imprint method "was quicker, simpler, and more efficient" in bacterial recollection.

REFERENCES

Campos-Murguía, Alejandro, et al. "Stethoscopes as potential intrahospital carriers of pathogenic microorganisms." *American Journal of Infection Control*, vol. 42, no. 1, 29 Oct. 2013, pp. 82-83, <https://doi.org/10.1016/j.ajic.2013.06.015>.