

Sponsored by:

solventum

Biofilm and Medical Devices: Implications for Sterile Processing and Infection Prevention

BY SYLVIE DUFRESNE, PH.D., MICROBIOLOGIST, SD CONSULTANT

This series of self-study lesson plans was developed by HSPA. Certified Registered Central Service Technician (CRCST) lessons provide members and certification holders with ongoing education on core Sterile Processing–related topics and processes. Purdue University's Extended Campus and HSPA both offer grading opportunities.

Earn Continuing Education Credits

Online: Visit www.myhspa.org for online grading.

By mail: For written grading of individual lessons, send completed quiz and $15 to:
Purdue Online – Central Service
Young Hall, Room 448
155 S. Grant St.
West Lafayette, IN 47907

Scoring: Each quiz with a passing score is worth two continuing education (CE) credits toward your CRCST recertification (12 credits).

Subscription series: Purdue Extended Campus offers an annual mail-in or online self-study lesson subscription for $75 (six specific lessons worth two credits each toward CRCST recertification of 12 credits). Call 765.494.0959 for details.

More information: HSPA provides online grading service for any of the Lesson Plan varieties. **Note: Purdue University provides grading services ONLY for CRCST and CIS lessons. Please do not send the CHL or CER lessons to Purdue for grading.** Direct any questions about online grading to HSPA at 312.440.0078. Questions about written grading are answered by Purdue University at 765.494.0959.

For paper/pencil scoring requests, please allow up to two weeks for grading.

LEARNING OBJECTIVES

1. Understand biofilm formation and its mechanisms of resistance
2. Identify where and how reusable medical devices become contaminated during use and processing
3. Recognize methods used to detect biofilm and assess cleaning effectiveness in medical devices
4. Understand strategies to prevent and remove biofilm through proper cleaning, disinfection, sterilization and monitoring

Biofilm plays a critical role in healthcare-associated infections. Microorganisms released from biofilm may infect patients directly or indirectly during device use and contribute to the transmission of pathogenic microorganisms in healthcare settings. Controlling biofilm is essential because it can interfere with the sterilization of medical instruments.

To ensure effective sterilization, it is necessary to monitor and validate both the initial cleaning process, which must remove biofilm, and the sterilization process itself. Monitoring helps ensure the safety of reused instruments and prevents associated infections. Recognizing the role of medical device reprocessing activities in preventing biofilm formation is fundamental to Infection Prevention and Control (IP&C) programs.

Objective 1: Understand biofilm formation and its mechanisms of resistance

Biofilm is a thin layer of microorganisms that adheres to a surface and becomes embedded within a self-produced extracellular polymeric substance (EPS) matrix, which also contains extracellular molecules and water channels (see **Figure 1**). EPS facilitates nutrient cycling, cell communication using chemical molecules, and the exchange of genetic material that enables the transfer of genes associated with antibiotic resistance. It also protects against host immune defenses, antimicrobial agents, such as disinfectants and antibiotics, and drying.[1]

Biofilm is composed of 90% water. Biofilm dry weight consists mainly of microbial cells (10-25%) and EPS (75-90%). This protective matrix acts as a barrier to the penetration of disinfecting and sterilizing agents, reinforcing the importance of effective cleaning

before sterilization. This highlights the importance of combining cleaning with sterilization process validation and monitoring to ensure adequate sterilization agent contact under defined conditions.

- Once established, biofilm exhibits significantly greater resistance to disinfectants, antibiotics, sterilization processes, and host immune defenses compared with free-floating microorganisms. This resistance is associated with multiple mechanisms that work together to protect the microbial community (see **Figure 2**).[2] Other critical aspects of biofilm include:
- Limited penetration of antimicrobial agents through the biofilm matrix – Antimicrobial compounds, whether antibiotics or chemical disinfectants, struggle to diffuse through the EPS structure at effective concentrations. This gradient of antimicrobial concentration means that only microorganisms at the biofilm surface experience lethal doses, while those deeper within the matrix are shielded from harm.
- Reduced metabolic activity of microorganisms – Many antimicrobial agents are most effective against actively growing cells that require active protein synthesis and cell wall turnover.
- Genetic adaptation – The biofilm environment selects variants that express enhanced resistance mechanisms, creating populations that are evolutionarily adapted to survive antimicrobial challenges.
- Presence of highly resistant microorganisms such as bacterial spores – These specialized structures possess thick protective coatings and extremely low metabolic rates, making them nearly impervious to most antimicrobial treatments. Because of this extreme resistance, bacterial spores are used in biological indicators to represent a worst-case microbial challenge for sterilization processes.
- Quorum sensing – Through the production and detection of signaling molecules, microorganisms within biofilm can activate shared survival strategies, including the production

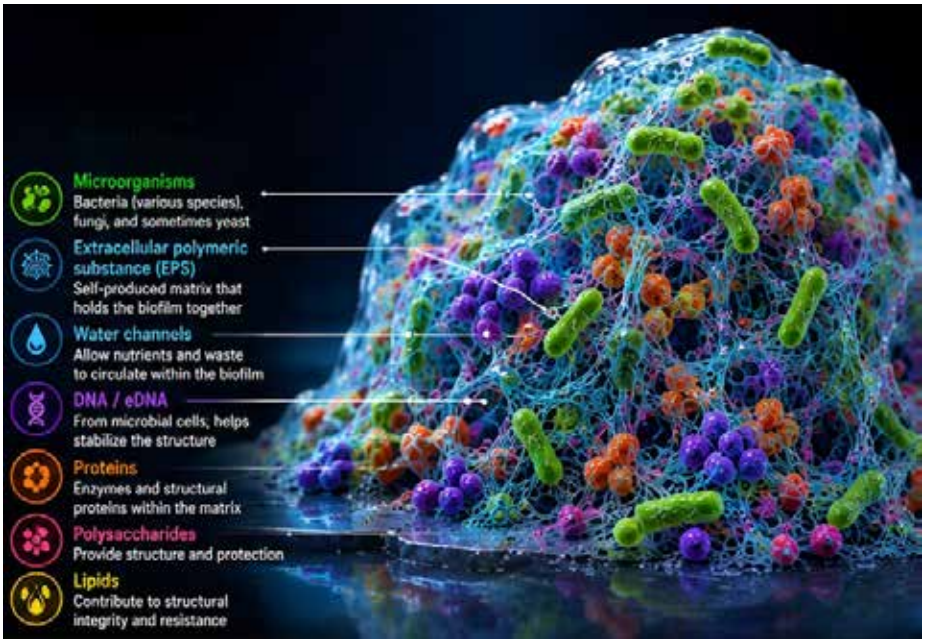


Figure 1. Biofilm composition (© SD consultant - Image generated by AI)

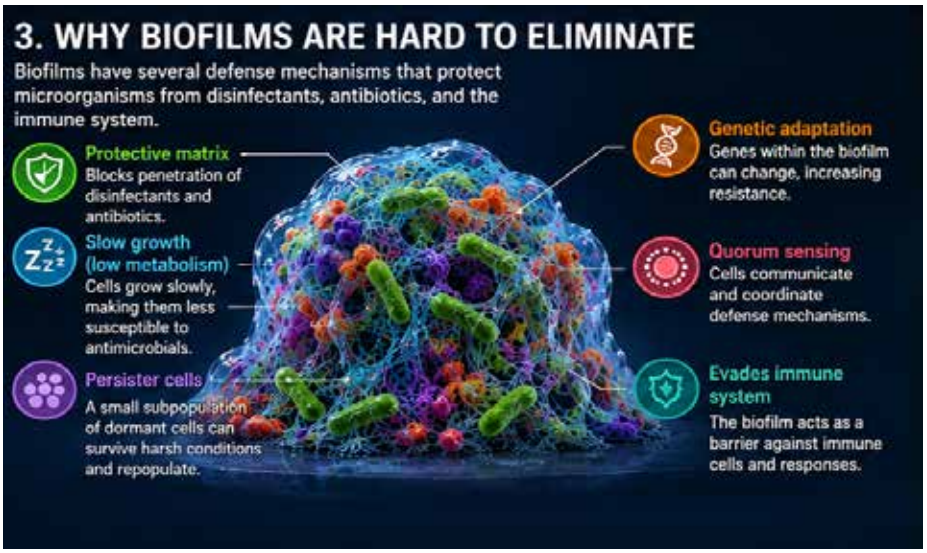


Figure 2: Biofilm defense mechanisms (© SD consultant – Image generated by AI)

of protective compounds and efflux pumps that actively remove antimicrobial agents. This coordinated response amplifies the protective effects of a formidable defense system that makes biofilm-associated infections particularly challenging to treat.

Understanding these mechanisms is essential for developing effective cleaning, disinfection and sterilization strategies for reusable medical devices.

Objective 2: Identify where and how reusable medical devices become contaminated during use and reprocessing

Understanding the sources of contamination is essential for implementing effective prevention strategies and maintaining proper infection control standards. Because these real-world contamination sources introduce variability that cannot be fully controlled, it is critical to reinforce the importance of cleaning and routine sterilization monitoring tools and practices.

Medical devices become contaminated through various pathways in the healthcare environment. Direct contact with a patient's bodily fluids, tissues and skin is a primary source of contamination. Healthcare personnel also contribute to medical device contamination when they handle devices. Environmental factors also play a major role, as contamination can occur from airborne and surface microorganisms.

Medical device design significantly impacts contamination risk. Poorly designed devices with complex features or difficult-to-access areas create challenges for proper cleaning and disinfection. A notable example includes the duodenoscope elevator mechanism, which has been associated with contamination incidents.[3]

Cleaning medical devices involves tap water, which contains microorganisms. The splash zone around sinks is particularly problematic, as water splashing during use can contaminate nearby surfaces and medical devices. Contaminated filters used for final rinse water in automated endoscope reprocessors (AERs) represent another critical concern. These filters can harbor microorganisms that may recontaminate devices during the final rinse cycle if not properly maintained or replaced according to manufacturer specifications.[4]

Inadequate, complex, long or multi-step manufacturer processing instructions represent a greater challenge that may lead to deviations from validated methods and reprocessing failures.

One advantage of sterilization processes over high-level disinfectants and liquid chemical sterilants is the use of sterilization monitoring tools designed to evaluate whether defined process parameters were achieved. Sterilization monitoring tools rely on defined, reproducible challenges rather than actual device contamination to evaluate process effectiveness. Still, this may not fully replicate the challenges posed by complex device designs if cleaning and preparation steps were not performed adequately or consistently. Thus, it is important to follow the manufacturer's instructions for use (IFU), as well as processing guidance from organizations such as the Association of periOperative Registered Nurses (AORN), the Association for the Advancement of Medical Instrumentation (AAMI), or the Operating Room Nurses Association of Canada (ORNAC).

Inadequate drying processes during medical device processing leave residual moisture that promotes microbial growth. Medical device storage requires particular attention to moisture control, as microorganisms thrive in wet or moist environments.

All disinfected medical devices must be dried thoroughly before storage to prevent microbial proliferation and ensure device integrity.[5] Sterilization processes using steam as the sterilizing agent have a drying stage, whereas low-temperature sterilization processes use deep vacuum to remove humidity from the packaged devices, effectively drying them. Devices should be stored in designated areas protected from dust, moisture and potential contamination sources.

Objective 3: Recognize methods used to detect biofilm and assess cleaning effectiveness in medical devices

Detecting biofilm on medical devices can be challenging because biofilm is often microscopic and not visible to the naked eye. **Table 1** presents indirect approaches used to detect biofilm alongside extraction methods, such as brushing or flushing lumens, swabbing surfaces, or directly inspecting medical devices with or without additional equipment.

Because none of the methods are perfect, multiple methods are often needed to verify whether biofilm is present on medical devices. These detection methods evaluate the presence of contamination or residual organic material but do not verify the effectiveness of disinfection or sterilization processes.

For this reason, cleaning verification and biofilm detection methods complement, but are distinct from, sterilization monitoring practices [e.g., biological and chemical indicators (BIs and CIs)]. Biofilm detection can be

Approaches	Methods	Advantages and/or Disadvantages
On extract	Microbiological methods (culture of the microorganism and identification)	· Detection levels can be low if extraction methods are not validated for biofilm removal or sampling · Can only detect the microorganisms that grow on the growth medium used
	Staining with crystal violet	· Detects most living bacteria, but no identification · Semi-quantitative
	ATP consumption measurement or fluorescent molecule detection (cleaning verification)	· Difficult quantification, high variability between bacteria, may detect inactivated bacteria
	Proteins (cleaning verification)	· Proteins may indicate other contamination, and not belong solely to the microorganism
In-situ	Visual inspection	· Hard to know if observation is due to a biofilm or physical damages
	Borescopes for channels	· Direct view inside channels · Limited by diameter
	Advanced biofilm detection microscopy with specific fluorescent markers	· Real-time visualization of biofilm presence and distribution · Only available for visually accessible surfaces

Table 1: Biofilm detection and cleaning verification methods

performed after cleaning during visual and functional inspection and before disinfection or sterilization. It can also be part of a medical device monitoring program when more complex testing is required, or when visual inspection is performed after disinfection to prevent recontamination of the medical device. Performing these assessments before sterilization is important, as sterilization monitoring tools cannot detect residual contamination present before the cycle.

Objective 4: Understand strategies to prevent and remove biofilm through proper cleaning, disinfection, sterilization and monitoring

Experts have identified three primary strategies to address biofilm-related problems in healthcare settings: 1. Preventing initial microbial attachment to surfaces, 2. inhibiting biofilm formation, and 3. eliminating or disrupting established biofilm. These strategies are supported by monitoring practices that verify process performance and help ensure that processing conditions are consistently achieved.

Preventing initial microbial attachment involves multiple strategies across different levels. Many mechanisms to prevent microbial attachment fall under medical device manufacturers' responsibilities, such as surface modification, antimicrobial materials or specialized coatings designed to inhibit colonization.

The healthcare setting plays an important role in limiting medical device contamination by preventing biofilm formation in processing equipment and designated areas through periodic disinfection and rigorous equipment maintenance. A critical preventive measure is to clean medical devices as soon as possible after use to remove organic material that may support microorganism attachment to surfaces and subsequent biofilm formation.

Drying after cleaning is also critical because humidity creates an environment favorable for microbial growth and biofilm formation. Additionally, healthcare professionals should be aware that silicone-based antifoaming agents, such as simethicone in gastrointestinal endoscopy, should not be applied directly through reusable medical devices like endoscopes. Because these agents are water-insoluble, they cause buildup on equipment and promote microbial biofilm formation.

Eliminating established biofilm is much harder than preventing its formation. Effective biofilm destruction typically requires a combination of mechanical action, chemical treatment, and validated disinfection or sterilization processes. Mechanical cleaning is particularly important because physical friction helps disrupt the biofilm matrix and remove embedded microorganisms. Enzymatic detergents, oxidizing agents, and other specialized cleaning chemistries may enhance biofilm removal when used as directed by the manufacturer. High-level disinfection and sterilization processes can further reduce microbial contamination, although their effectiveness may be compromised

if biofilm is not adequately removed during cleaning.

Although BIs and CIs confirm that process conditions were achieved, their results are based on standardized challenges and do not account for residual biofilm or soil that may remain on device surfaces. However, monitoring provides documented evidence that the sterilization steps comply with standards, reducing the risk of failure.

Conclusion

Prevention remains the most effective strategy for biofilm management. Healthcare settings must implement rigorous device-cleaning protocols, establish proper environmental controls in processing areas, maintain equipment in compliance with manufacturer guidelines, and ensure appropriate storage conditions. By addressing these contamination sources systematically, healthcare organizations can significantly reduce biofilm formation and infection risks and improve patient safety outcomes.

Preventive measures include immediate pretreatment at the point of use, thorough manual and automated cleaning, adherence to validated processing instructions, and routine monitoring practices using cleaning verification methods and BIs and CIs to confirm sterilization process performance. These monitoring tools verify that defined conditions have been met, but they do not detect residual biofilm or contamination, reinforcing the need for a comprehensive, multi-step approach to device processing. P

REFERENCES

1. Sharma, S., Mohler, J., Mahajan, S. D., Schwartz, S. A., Bruggemann, L., & Aalinkeel, R. (2023). Microbial Biofilm: A Review on Formation, Infection, Antibiotic Resistance, Control Measures, and Innovative Treatment. *Microorganisms*, *11*(6), 1614.
2. Maillard JY, Centeleghe I. (2023). How biofilm changes our understanding of cleaning and disinfection. *Antimicrob Resist Infect Control*. 2023 Sep 7;12(1):95.
3. Humphries RM, McDonnell G. Superbugs on Duodenoscopes: the Challenge of Cleaning and Disinfection of Reusable Devices. *J Clin Microbiol*. 2015 Oct;53(10):3118-25.
4. Walker JT, Bak A, Marsden G, Spencer W, Griffiths H, Stanton GA, Williams C, White LJ, Ross E, Sjogren G, Bradley CR, Garvey M. (2022). Final rinse water quality for flexible endoscopy to minimize the risk of post-endoscopic infection. Report from Healthcare Infection Society Working Party. *J Hosp Infect*. Jun;124:79-96.
5. Alfa MJ, Singh H. (2020) Impact of wet storage and other factors on biofilm formation and contamination of patient-ready endoscopes: a narrative review. *Gastrointest Endosc*. Feb;91(2):236-247

CRCST Self-Study Lesson Plan Quiz: Biofilm and medical devices: Implications for Sterile Processing and Infection Prevention

Lesson No. CRCST 211 (Technical Continuing Education – TCE) · Lesson expires December 2029

1. Which best defines biofilm?
   a. A layer of free-floating cells
   b. A thin layer of microorganisms adhering to a surface
   c. A type of protective human tissue
   d. A layer of liquid on medical devices

2. An extracellular polymeric matrix (EPS):
   a. Aids antibiotic penetration
   b. Nourishes a single type of bacteria
   c. Protects microorganisms from immune defenses and antimicrobials
   d. Separates biofilm cells

3. Biofilm is more resistant to antimicrobial treatments.
   a. True
   b. False

4. Which is a common source of device contamination?
   a. Device handling
   b. Environmental factors
   c. Direct contact with a patient's bodily fluids
   d. All the above

5. How does medical device design influence contamination risks?
   a. Stainless steel resists bioburden
   b. Devices with hard-to-reach areas complicate cleaning and drying and promote biofilm
   c. Biofilm grows faster on plastic
   d. Lumened devices always harbor hardened biofilm

6. Which practice most effectively prevents biofilm formation?
   a. Allowing devices to dry slowly at room temperature
   b. Cleaning and drying devices as soon as possible after use
   c. Flushing devices with saline
   d. Storing devices in a warm room

7. Why are silicone-based antifoaming agents, such as simethicone, not recommended for reusable devices?
   a. They are expensive
   b. They cause corrosion
   c. They promote biofilm formation and accumulation
   d. All the above

8. What is the most effective way to remove established biofilm?
   a. Soaking devices in hot water
   b. Using a validated combination of mechanical cleaning, chemical treatment, and disinfection/sterilization
   c. Air-drying devices for at least 24 hours
   d. Scrubbing the device with saline and an approved brush

9. What is a key limitation of chemical and biological indicators (CIs/BIs)?
   a. They cannot be used in routine monitoring
   b. They can only detect blood
   c. They do not detect residual soil or biofilm on devices
   d. None of the above

10. What is the main difference between biofilm detection methods and sterilization monitoring tools?
   a. Detection methods identify contamination; monitoring tools verify process performance
   b. Detection methods verify sterilization conditions; monitoring tools aid decontamination
   c. Monitoring tools are only used before cleaning
   d. Detection methods are required by the Joint Commission

11. A sterilization cycle with acceptable CI and BI results indicates:
   a. The device is free of all contamination, including biofilm
   b. The sterilization process met defined conditions
   c. The device was properly cleaned
   d. Recontamination is unlikely after the cycle

12. Extended soak times and isolation are effective for addressing biofilm.
   a. True
   b. False

13. Biofilm detection can be performed:
   a. With the naked eye
   b. Before cleaning and during visual and functional inspection
   c. After cleaning, during visual and functional inspection, and before disinfection or sterilization
   d. Only by experienced technicians

14. Biofilm is comprised of:
   a. Mostly fats and oils
   b. 90% water
   c. 90% proteins
   d. Millions of resistant spores

15. Enzymatic detergents and oxidizing agents:
   a. May contribute to biofilm formation
   b. May enhance biofilm removal
   c. Are only appropriate for non-lumened devices
   d. Cause corrosion

Request for paper/pencil scoring

(Please print or type information)

❍ I have enclosed the scoring fee of $15. (Please make checks payable to Purdue University. We regret that no refunds can be given.)

❍ Check here if you have a change of address.

❍ Check here if you wish to have your results emailed to you.

Detach quiz, fold and return to:
Purdue Online – Central Service
Young Hall, Room 448
155 S. Grant St.
West Lafayette, IN 47907

For paper/pencil scoring requests, please allow up to two weeks for grading.

Name

Mailing Address (Be sure to include apartment number or post office box.)

City | State/Province | Zip Code/Postal Code

Daytime Telephone | HSPA Membership Number

Email Address

If your name has changed in the last 12 months, please provide your former name. Purdue University is an equal access/equal opportunity institution.