Disinfection Guideline for Cryostats
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If your laboratory is inspected and accredited by the College of American Pathologists, please refer to the most current requirements. According to the 2018 CAP accreditation checklist, “The cryostat must be defrosted and decontaminated by wiping all exposed surfaces with a tuberculocidal disinfectant. The cryostat should be at room temperature during decontamination unless otherwise specified by the manufacturer. This should be done at an interval appropriate for the institution; this must be weekly for instruments used daily. Trimmings and sections of tissue that accumulate inside the cryostat must be removed during decontamination. Although not a requirement, cut-resistant gloves should be worn when changing knife blades.”

Wear Personal Protective Equipment (PPE):
Personal Protective Equipment, such as gowns, puncture and penetration resistant gloves and eye protection must be worn when performing cryostat disinfection procedures.

Cryostat Preparation
- Remove used blades/knives from their holder.
- Dispose of blades according to the policies and procedures of the institution. Disinfect knives by soaking them in a non-corrosive tuberculocidal disinfectant solution.
- Remove all sectioning debris, utensils, tools and accessories from the cryochamber.
- Sectioning debris must be removed because organic material (blood and proteins) will prevent access to contaminated surfaces and may contain high concentrations of microorganisms, which could possibly inactivate the disinfectant. All sectioning debris should be treated as biohazardous waste and disposed of according to the policies and procedures of the institution.
- Utensils and tools (pencils, forceps, brushes, etc) are removed so they do not block access to the cryochamber walls or microtome surfaces. They should be soaked in the disinfectant solution for the appropriate time, rinsed in water and allowed to dry.
- Accessories should be placed in the non-corrosive tuberculocidal disinfectant solution for the recommended exposure time, rinsed with water followed by absolute alcohol and then allowed to thoroughly dry on absorbent toweling.
- Accessories with multiple components should be disassembled to ensure proper disinfection.
- Follow the manufacturers recommendations for lubrication before exposing to cold temperatures.
- Ethyl, reagent or isopropyl alcohol can be liberally applied to clean the cryochamber and provide disinfection. These alcohols are rapidly bactericidal, tuberculocidal, fungicidal, and virucidal, but do not destroy bacterial spores. Their cidal activity drops sharply when diluted below 50% concentration, and the optimum bactericidal concentration is between 60%–90% solutions in water (v/v). Absolute alcohol is less bactericidal than lower concentrations because proteins are denatured more rapidly in the presence of water *.
- If desired, application of absolute alcohol can follow the lower concentrations to remove any residual water.

*https://www.cdc.gov/infectioncontrol/guidelines/disinfection/disinfection-methods/chemical.html

Integrated Disinfection Systems
If available, integrated disinfection systems installed by the cryostat manufacturer can be activated for the time recommended by the manufacturer.

- To assure that the system is capable of complete and thorough disinfection of the entire cryochamber and microtome, request written documentation regarding the testing procedures used, efficacy of the system against all tested pathogens, amount of time required for complete disinfection for specific pathogens and information regarding any areas within the cryostat that will not be properly disinfected.
- If there is not total confidence in the thoroughness of the integrated system, chemical disinfection must follow.
Chemical Disinfection

To disinfect a cryostat using a chemical disinfectant, the instrument MUST be at room temperature before the process is started.

- Turn off and unplug the instrument before beginning the disinfection process.
  - Once the cryostat has reached room temperature, do not turn the handwheel until it has been returned to cold operating temperature.
  - Do not use a hair dryer in the cryochamber to accelerate the warming process because this will create aerosols.
- Do not create aerosols by spraying disinfectant (or anything else) in the cryostat chamber. Use a non-corrosive tuberculocidal disinfectant to avoid damage to the cryostat and its accessories.
- The EPA maintains a list of Antimicrobial Chemical/Registration Number Indexes on their website and it is updated regularly. From the links below you can find agents effective against bloodborne pathogens such as Mycobacterium tuberculosis, HIV-1 virus, and Hepatitis B or Hepatitis C virus and others.
  - It is critical to remember that NONE of these solutions have been tested at low temperatures and can only be used at room temperature.
- Pour disinfectants onto absorbent disposable toweling placed on the cryochamber walls and exposed microtome surfaces. Allow them to remain in contact with contaminated surfaces for the length of time specified in the disinfectant's instructions.
- Any disposable material used in the disinfection process must be disposed of in accordance with the policies and procedures of the institution.

Following Disinfection

After the disinfection procedure is complete, the cryostat and all accessories must be thoroughly dried and lubricated, if specified by the manufacturer, before being put back into service at cold temperatures.

- Absolute ethyl alcohol can be used to remove residual moisture from surfaces. Accessories with multiple parts, such as the disposable blade holder, knife holder and their respective bases, must be thoroughly dried before reassembly. Disinfected utensils must also be thoroughly dried.
- Plug the instrument in and turn it on. Do not turn the handwheel until the microtome has reached cold temperature.
- Lightly lubricate only the areas specified by the manufacturer using only their recommended lubricants.
- The liquid waste container should be emptied in accordance with the policies and procedures of the institution.
- Before replacing, add a small amount of liquid bleach to the empty container. Household bleach solutions contain 5.25% sodium hypochlorite, but effective disinfectant working solutions should 0.5%-2% sodium hypochlorite. Discard the waste once the volume reaches approximately 10 times the volume of the original amount of bleach.

For optimum sectioning, allow the instrument to cool long enough for the metal microtome parts to reach the cold temperature setting. Consult the Instructions for Use Manual of the cryostat to determine the approximate time required. Most cryostats require no less than 3 hours.

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This document is intended to serve as a guideline ONLY and NOT AN absolute recommendation for cryostat decontamination. Each laboratory is advised to use these guidelines as a starting point and modify certain parameters to fit state and local institutional requirements and laws, as appropriate. The use of the information contained in this guideline does not guarantee compliance with the CAP accreditation requirements or regulations from other accrediting organizations. Some information may be different or more stringent than the published CAP Checklists.