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## Sterilizer (Consolidated) V.2

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### ABSTRACT

How to use a Sterilizer/Autoclave (general) for the Bennett Lab.

### MATERIALS

Personal Protective Equipment:

- Dry arm-length autoclave gloves
- Safety glasses
- Closed-toe shoes
- Pants
- Neoprene overcoat or lab coat (optional)
- Face shield (optional)

## SAFETY WARNINGS



Users **MUST** receive training, read and understand the operating manual for this piece of equipment prior to use. **Personal Protective Equipment required.**

**Specific Hazards:** Hot Surfaces, Hot Liquids.

### Prepare Autoclave

- 1 Sign up in advance to use the autoclave.  
Turn on autoclave.
- 2 Make sure that the sterilizer chamber as well as the chamber drain strainer (inside the drain hole near the sterilizer door) is free of any debris. **This should be done before every cycle, every day.**  
A clean drain strainer is essential for safe operation of the sterilizer.
- 3 Press the "Jacket" button, to get the jacket pressure up so that the autoclave cycle will take less time.

### Prepare Materials (Liquid or Solid) and Load Autoclave

- 4 Appropriate secondary containers are stainless **steel** or **aluminum trays** and **Nalgene bins**.  
Choose the deeper Nalgene bins if you are autoclaving tall items, though you may need to add ~1" of roH<sub>2</sub>O to ensure even distribution of heat.
- 5 Label items with autoclave tape, to indicate that the item has been autoclaved.
  - 5.1 **You absolutely must use secondary containment if autoclaving media with AGAR or anything else that might make a mess in the chamber (e.g. bags of soil, used media plates, potentially heat-labile plastic).**
  - 5.2 Secondary containment is not necessary for other items but may help in moving multiple

smaller items in and out of the autoclave more easily.

- 5.3** Be certain that any plastics you are sterilizing are autoclavable. **If you are still uncertain, use secondary containment. Do not hesitate to ask questions.**

- 6** Load chamber appropriately, **wearing dry orange autoclave gloves** to protect your hands.

**CONSIDER EASE AND SAFETY WHEN LOADING AND REMOVING ITEMS into the chamber. For example, do not load tall flasks with liquids on the top shelf where there is little space.** Remove the shelf if you are sterilizing extra-tall items.

## Liquid Cycle

- 7** Do not sterilize liquid media in the same secondary container as dry material. Liquids may boil over during the cycle and wet the dry material.

- 8** **Loosen any screw caps** before starting sterilization. If caps are tight, pressure may build inside the container, risking **explosion** or **container deformation**.

- 9** **Never sterilize any volatile chemicals, pesticides, alcohols, bleach, chlorides, or any hazardous materials!**



## Solid Cycle

- 10** Biohazardous waste bags should be zip-tied to allow for air escape.

- 10.1** The bag should be rotated to have the opening at the bottom to allow for any liquid to drain into the bins and not into the autoclave.

## Check Autoclave Settings

- 11 Close the door and turn the wheel approximately 6 times from the completely open position. Steam will exit from the door if not tightened sufficiently. Tighten securely but not excessively; **overtightening the wheel will damage the door.**
- 12 The temperature should be set at 250°F for most applications. If it is not at 250°F or you need to use a different temperature, ask for assistance.
- 13 Conserve water and energy by utilizing as much autoclave capacity whenever possible. Plan ahead and check with others about autoclaving needs before turning on the machine. **However, do not compromise SAFETY.**
- 14 If sterilizing agar media, check that the “Fluid” setting is on. This ensures that the chamber pressure will be reduced very slowly in order to minimize boil-over of liquids. If you wish to dry your items, the “Dry” cycle may be used.
- 15 Check that the autoclave is set for 20 minutes. This is sufficient time to sterilize most materials, so use 20 minutes unless you’re given specific instructions to use another duration.
- 16 Press RUN, 01, and ENTER to start the autoclave. Set your personal timer to ~50 minutes if you are doing a fluid cycle because it will take approximately that much time for the autoclave to return to atmospheric pressure. Set your timer 90 minutes if you need a drying cycle. It is a good idea to check back after 5-10 minutes to make sure that the autoclave door is completely shut.

## Removing Items from Autoclave and Shutting Down

- 17 The monitor screen will indicate when the sterilization cycle is finished. You should be wearing closed-toe shoes. **Put on the Personal Protective Equipment** on the cart or hanging on the cart or the wall by the autoclave: **arm-length DRY autoclave gloves, and neoprene apron or lab coat.**
- 18 **Open door slowly and only partly. Stand away from the autoclave** and allow a few seconds for steam to escape before opening completely.

- 18.1** If water leaks from the front of the sterilizer, DO NOT open the door. Contact service personnel.
- 19** Carefully remove sterilized materials and place onto a cart to cool. Move the cart back to the lab and use the "HOT" sign to warn lab mates from burning themselves.
- 20** If you need to run another cycle and the monitor has turned off, switch #16 in the panel box off and back on. **If the sterilizer will not be operated again during the day, close the green water valve in the back of the autoclave, turn off switch #16 in the panel box, and pull down the handle on the breaker box.**
- 21** If there have been any spills or if any debris has littered the sterilizer, allow autoclave to cool fully before cleaning. Clean it thoroughly with water, sponges, scouring pads, and paper towels before further use. Never use bleach or any other chlorides to clean the sterilizer – these are corrosive and will damage the autoclave.