

**Standard Operating Procedures (SOP)
Autoclave Usage and Disposal of Waste**

Autoclave Make/Model: Priorclave 150L/320L



Located in Building/Room: DBH 143 and MH 385

Purpose

To provide general guidance in the use of autoclaves in Dan Black Hall and McCarthy Hall. Autoclaving is used to destroy microorganisms that are found in Biosafety Level 1 (BSL1) waste and sterilization of microbiological equipment used at Biosafety Level 1 and 2 Laboratories at CSUF.

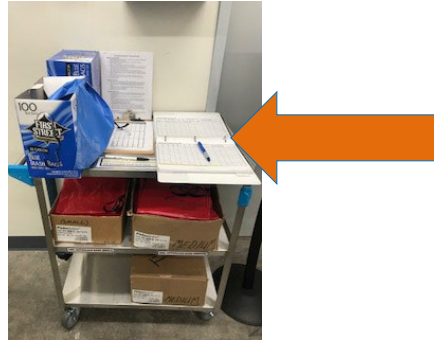
Potential Hazards

1. Heat burns from hot materials and autoclave chamber walls and door.
2. Steam burns from residual steam coming out from autoclave chamber and materials on completion of cycle.
3. Hot fluid scalds from boiling liquids and spillage in autoclave.
4. Hand and arm injuries when closing the door.
5. Body injury if there is an explosion.
6. Toxic fumes from materials that are not compatible with autoclaving process.

PPE

1. Lab Coat
2. Safety Goggles
3. Closed-Toe Shoes
4. General Lab Gloves (e.g. Nitrile Gloves)
5. Heat Resistant Gloves (Autoclave Gloves)
 - a. Caution: Autoclave gloves will absorb liquids that conduct heat energy, which may cause burns to the hands.
6. Face Shield
 - a. Recommended when removing liquid loads.

3. The log book must be kept on the autoclave supply cart at all times.



Procedures for Autoclaving Waste

*****NOTE: THIS SOP IS APPLICABLE TO BSL1 LAB WASTE ONLY! Waste originating from BSL2 Laboratories is not allowed to be autoclaved and disposed in a municipal dumpster or sanitary sewer w/o prior approval from PI or Biosafety Officer.**

1. Preparing Material for Autoclaving
 - a. Ensure material is safe for autoclaving (e.g. plastics that can withstand the temperature setpoint in the program that is chosen.)
 - b. NEVER AUTOCLAVE FLAMMABLE, REACTIVE, CORROSIVE, TOXIC, or RADIOACTIVE MATERIALS.
 - c. Glassware must be inspected for cracks prior to autoclaving.
 - d. Prepare and package material suitably. Waste loads should have ½ cup (100 ml) of tap water added to the inside of autoclavable red biohazard bags. Do not fill a waste bag over half full or above its fill line (if present.)



- e. Loosely encircle the neck of red biohazard bags with autoclave tape, leaving a hole approximately the size of your first to allow steam to penetrate and pressure to escape.
- f. Bottles and liquid containers should never be tightly sealed. Do not fill Erlenmeyer flasks more than $\frac{1}{2}$ full.
- g. All items must be placed in the provided metal secondary containers (discard trays) to secure the contents and contain potential spills. Before loading items into the discard trays, make sure to pour any liquids that might be hiding beneath the perforated false floors into the room's sink while running plenty of water from the faucet to keep the sink clean.
 - i. If tray contains agar, then agar must be allowed to solidify and scraped into designated red step-on receptacle ONLY. Agar should never go down the sink! See Step 8 (Maintenance) for additional information.



- h. Dry waste loads should never contain glass, sharps, or any other materials that might cut or puncture the red autoclave bag.
 - i. Waste loads with agar plates must be run on a liquids program.
2. Loading an Autoclave
- a. Check the log book to see what autoclave is available
 - b. Wear the required PPE
 - c. Open the door
 - i. Door should be ajar from the previous user. Pull the yellow handle towards you to open. If the door is closed, see page 6.

- d. Place material in autoclave (Recommend using cart to transfer material and associated spill tray to autoclave.) Do not overload; leave sufficient room for steam circulation. Waste bags should be placed in a vertical orientation to avoid spills.



- e. Close the door and latch it completely by following the pattern on the door.

i.



3. Operating Autoclave

- a. Choose the appropriate program for the container and material being sterilized. Consult the autoclave program chart located on autoclave door for assistance in choosing the appropriate cycle.

| Program Number | Cycle Type | SAFE FOR PLASTIC | Hold Temp (°C) | LOAD TYPE OR LIQUID VOLUME PER FLASK | Can I Run Waste? | Hold Time At Temp (min) | Cycle Time (min) |
|----------------|------------|------------------|----------------|---|------------------|-------------------------|------------------|
| 1 & 2 | Gravity | YES | 121 | ≤ 250 ml 250 - 500 ml | NO | 30 | |
| 2 & 7 | Gravity | NO/ GLASS ONLY | 123 | ≤ 250 ml 250 - 500 ml | NO | 20 25 | |
| 3 & 8 | Gravity | NO/ GLASS ONLY | 123 | 500 - 1000 ml 1000 - 1500 ml | NO | 30 35 | |
| 4 & 9 | Gravity | YES | 121 | 19 x 23 Bags of Waste (NO SEALED CONTAINERS in the Bag) | YES | 30 | |
| 5 & 10 | Vacuum | YES | 121 | Plastics (Pipette Tips) Erlenmeyer Flasks w/ Loose Foil Covers & Instruments | NO | 20 40 | |

- b. Only designated individuals are allowed to set and/or change parameters for the autoclaves.

- c. Start your cycle (PRESS START BUTTON) and fill out the autoclave user log with your contact information. A completed cycle takes a minimum of 75 minutes.



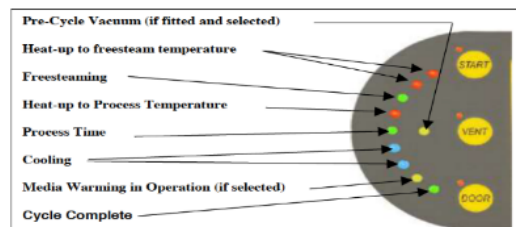
- d. Do not attempt to open the door while the autoclave is operating.
- e. If problems with the autoclave are perceived, abort the cycle by pressing the emergency stop button, place an “Autoclave Out of Order” sign on autoclave door, and report problem according to autoclave contact list that is posted in the room with the autoclaves. You will not be able to retrieve your items until someone with the appropriate key unlocks the emergency stop button and ensures that the interior is depressurized and cool enough to open the door.



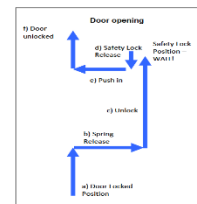
| DBH AUTOCLAVE CONTACT LIST | | |
|--|------------------|-------|
| Person | Phone (657)-278- | Email |
| | | |
| | | |
| AFTER HOURS (BEFORE 8AM, AFTER 5PM) AND ON WEEKENDS AND/OR HOLIDAYS SEND AN EMAIL TO: | | |
| | | |
| | | |

4. Unloading Autoclave

- a. Wear the required PPE. A face shield is recommended when removing liquid loads.
- b. When the cycle is complete (indicated by the final LED in the progress display semicircle being illuminated), open the door.



- c. Open the door
 - i. PRESS DOOR button – it will beep.
 - ii. HOLD will appear on the time/temp display.
 - iii. WAIT 20 seconds and then the HOLD will go away (it will beep again).
 - iv. Door indicator illuminates – press door button a second time and then open with the latch.



- d. Carefully unload the contents of the autoclave, making sure to avoid getting the autoclave gloves wet since they would then lose their insulating properties.
 - i. Recommend placing material and associated spill tray on cart located in room to allow for additional cooling if needed.



- e. Close the autoclave door but DO NOT LATCH IT. (The autoclave chamber can take up to 10 minutes to refill after a cycle is complete. If the door gets latched too soon after a run, the autoclave can get a fault condition that requires a special key to be reset.)
- f. Ensure that caps remain loosely secured to media bottles when removing and transporting liquids. To avoid possible implosion of media bottles caused by vacuum formation from cooling media, do not fully tighten caps until media bottles have cooled to near ambient temperature.

- g. Solid waste loads in red biohazard bags must be placed in a blue bag and disposed in an outside dumpster. (Make sure that the blue bag is firmly secured with a knot or a twist tie.) Liquid waste is to be poured down the autoclave room sink (i.e. a sanitary sewer drain) with plenty of water.



5. Equipment Malfunction

- a. If the autoclave does not operate exactly as expected, do not attempt to fix the problem.
- b. Abort the cycle by pressing the emergency stop button, place an “Autoclave Out of Order” sign on autoclave door, and report problem according to autoclave contact list that is posted in the room with the autoclaves. You will not be able to retrieve your items until someone with the appropriate key unlocks the emergency stop button and ensures that the interior is depressurized and cool enough to open the door.

6. Spill Response Procedures

- a. Spills may occur from a boil-over, a broken liquid container, or a torn waste bag.
- b. No operation of the autoclave is allowed until all spills are cleaned up.
- c. Wait until the autoclave and materials have cooled to room temperature before attempting spill cleanup.
- d. Non – agar spills contained in a metal tray should be taken to autoclave room sink and cleaned out using household dishwashing liquid (such as Dawn, Palmolive, or a “green” alternative) and a non-abrasive scrubbing device (such as a blue-colored non-scratch scour pad from Scotch-Brite.) Agar spills must be allowed to solidify and scraped into

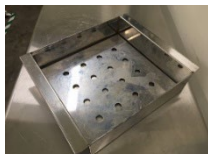
- e. designated red step-on receptacle ONLY. See Step 8 (Maintenance) for additional information. Do not use harsh detergents, bleach, abrasive cleansers, steel wool, or abrasive green-colored scour pad. Make sure that final rinse of the autoclave trays is with DI water so as to avoid the accumulation of mineral deposits from tap water.
- f. Spills not contained in a metal tray require an “Autoclave Out of Order” sign to be placed on the unit, pressing the emergency stop button to make the unit unavailable, and report the problem according to the autoclave contact list that is posted in the room with the autoclaves. In these cases, staff members will need to assess the nature/extent of the spill and then determine how best to clean the area under the bottom shelf of the autoclave.

7. Incident Response

- a. Incidents that result in injury or spill of material greater than 1 liter must be reported to your lab supervisor and Environmental Health and Safety at x7233 (SAFE).
- b. If an injury occurs, seek first aid. For medical assistance dial 911.
- c. If clothing is soaked, remove it and place the injury in cool water.
- d. Place an “Autoclave Out of Order” sign on the unit indicating that it is not to be used until it is safe for operation and press the emergency stop button to make the unit unavailable until the situation is resolved.

8. Maintenance

- a. Occasionally residual waste material (typically gelatin consistency; agar spills) located underneath the false floors of the discard trays will require removal by means of scraping. Waste is to be discarded into designated red step-on receptacle ONLY, which is located in the autoclave room.



- b. Submit a biological waste pickup request when receptacle becomes $\frac{3}{4}$ full by:
 - i. Completing request online:
 - ii. <https://ehs.fullerton.edu/services/>
 - iii. or call Environmental Health and Safety at x7233 (SAFE)

*****NOTE: Our campus autoclaves are regulated by the Orange County Health Care Agency. Failure to abide by this SOP could potentially lead to fines or violations. If you have any questions about autoclaves or autoclave related issues, please contact your department and/or EHS (x7233).**

Responsible Executive: Vice President for Administration and Finance

Responsible Office: Environmental Health and Safety

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