

## STANDARD OPERATING PROCEDURE

### Autoclaves

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Autoclaves use pressurized steam to destroy microorganisms.

Minimum Personal Protective Equipment Required: Heat insulating gloves, eye protection, lab coat, long pants, and closed-toe shoes.

Risks: Burns from touching a hot surface, scalding from steam

Special Handling:

- ✓ Use only autoclave safe plastics in the autoclave (polypropylene)
- ✓ Always use heat resistant gloves when operating autoclave or moving items to and from the autoclave.

Protocol/Procedure:

1. Place items to be autoclaved in an autoclave safe container (polypropylene tub). Never place glassware or bags directly on the bottom or floor of the autoclave.
2. Prepare items for autoclaving
  - a. For loose items (microcentrifuge tubes, spatulas, scissors, etc.) place in an autoclave safe container with a lid, a beaker covered in foil, or wrap the item(s) in foil.
  - b. Loosen any caps on bottles or containers.
  - c. If there are any stoppers on bottles or tubes, place a second upside-down autoclave bin on top of first bin to contain any stoppers that may pop out.
  - d. Containers of media or buffer should not be above the containers full line.
  - e. Containers of agar media should not be more than ½ full.
  - f. Do not crowd the tubs. There should be room for the steam to move around all items in the tub.
  - g. Place autoclave tape on the tub or individual items
  - h. *Biohazard waste:* Add one cup of water to each bag of solid waste and keep the bags open. Polypropylene biohazard bags are impervious to steam, so they need to remain open to allow steam to enter.
3. Transport autoclave tubs to autoclave on a cart. You will need to go through the freezer room to get to the autoclave room.
4. Press the DOOR button to open the autoclaves. Step back from the autoclave to avoid any steam or heat escaping from the autoclave.

5. Using heat resistant gloves, place the tubs into the middle of the autoclave.
6. Close the autoclave door by pressing and holding DOOR until the door is closed.
7. Make sure the display lights are lit for READY, DOORS CLOSED, and START.
8. Select the cycle to run.
  - a. Use liquid cycle (slow exhaust) when autoclaving liquids (including biohazard bags), to prevent contents from boiling over.
  - b. The gravity cycle (fast exhaust) should only be used when autoclaving dry items.
  - c. **If you have any doubts as to what cycle to use, use the liquid cycle.** It is fine to autoclave dry items with the liquid cycle.

Autoclave Cycles on all 3 Autoclaves					
Cycle Number	Cycle Type	Sterilize Temp (°C)	Sterilize Time (Min)	Dry Time	Use (Zhou lab)
1	Liquid	122	45	15	
2	Liquid	122	50	15	<b>Biohazard waste</b>
3	Gravity	122	35	20	<b>Glassware/plasticware, no liquids</b>
4	Gravity	122	45	20	
7	Liquid	122	30	15	<b>Media and buffers</b>

9. Press the number corresponding to the cycle you want. For cycle 7, press 4, up arrow, and 3. The number 7 should be shown on the display.
10. Press START. Make note of when the autoclave should complete the cycle.
11. Once complete, COMPLETE should be lit.
12. Wait a full five minutes for dry goods and ten minutes for liquids before removing items after the completion of a run.
13. Autoclaves should be kept closed at all times. When left open, they have constant pressure on the door cylinders which reduces the lifespan.
14. Report any suspected issues to the building manager right away.
15. All autoclave waste should be put in the black trash bags provided by SRTC in the autoclave room. The trash should be tied, and the lab is responsible for making sure it goes into the dumpster at the SRTC dock area. You can see Vanessa Smith in the Storeroom for more bags if needed.