Dahlquist Lab Last modified: 5/28/08

## **Autoclave Instructions**

1. Check the autoclave to see if it is turned on. If the autoclave is off, turn it on by pressing the "Power" switch behind the black door on the top left hand corner of the autoclave.

- 2. Make sure you are wearing gloves.
- 3. Prepare the items for autoclaving:
  - a. Media (or other solutions) should be prepared just before autoclaving so that contaminants do not have a chance to grow before the media is sterilized. Flasks should have aluminum foil covering the opening. Fold over the aluminum foil and make sure there is a generous amount covering the lid. The caps on bottles should be loosened.
  - b. Clean glassware should have aluminum foil covering the openings; the caps on bottles should be loosened. Clean spatulas should be completely wrapped in aluminum foil. Culture tubes should have metal caps.
  - c. Biohazard waste in bottles or flasks should have all tape removed. The caps on bottles should be loosened. Biohazard wasted in the biohazard waste bag should have the top twisted shut and closed with autoclave tape. The entire pot can go into the autoclave.
- 4. Place autoclave (temperature sensitive) tape on all items. Use the smallest piece of tape possible; at least one stripe should be visible on the piece of tape. The tape can go directly on foil lids. Small pieces of tape should be placed on bottle caps with one end folded over for easy removal. Avoid putting autoclave tape on the glass itself.
- 5. Place materials to be autoclaved into a plastic tray. This is especially important for autoclaving liquids. The tray will catch the spill if the liquid boils over during the cycle. Biohazard waste bags can be autoclaved directly in the steel pot. Dry items can be placed on a metal cafeteria tray instead of the larger plastic tray. Be sure to check whether the tray you are using is autoclave safe (i.e., won't melt in the autoclave). If you are not sure, ask.
- 6. Open the autoclave door by turning the wheel handle to the left ("lefty-loosey").
- 7. Place the tray inside the autoclave, being careful not to burn yourself on the sides of the chamber.
- 8. Close the door by applying even pressure to the door and turning the wheel handle to the right ("righty-tighty") until the indication that reads "door unlocked" is gone. Turn the wheel as far as it will go without exerting undue force.
- 9. Select the appropriate cycle, either liquid or gravity by pressing the button twice:
  - a. When autoclaving liquids, **ALWAYS** select a liquid cycle. The liquid cycle releases the pressure slowly over a 20 minute period after the sterilization period is finished so that the liquids do not boil over when the pressure is released.
  - b. Use the gravity cycle for dry glassware.
  - c. 1 = liquid: 15 minute sterilization followed by 20 minute slow exhaust
    - 2 = liquid: 20 minute sterilization followed by 20 minute slow exhaust
    - 3 = gravity: xx minute sterilization cycle, fast exhaust, followed by 20 minute drying cycle
    - 4 = liquid: xx minute sterilization followed by 20 minute slow exhaust
- 10. Wait until the temperature rises up to 121°C and the pressure reaches 20 psi (the sterilization timer will begin once these parameters are reached).
- 11. Sign the log book with your name, the lab name, date, time, and cycle used.
- 12. Take note of the time it will take for the cycle to be completed plus additional exhaust or drying time, then set a timer to remind you when the autoclave cycle is finished.
- 13. When the cycle is completed, carefully open the autoclave door by turning the wheel handle to the left and open the door slowly, just a crack to let the steam escape. Be careful so that you do not get burned by the steam.
- 14. Wait ten minutes before removing the items from the autoclave. Be sure to use the terry cloth gloves because the items and the chamber will be extremely hot.
- 15. Wait for the items to cool down to room temperature before tightening bottle lids and putting the items away.
- 16. If you are the last one using the autoclave for the day, put the autoclave in standby mode by pushing the "Standby/Off" switch behind the black door on the upper left side of the autoclave.