Tonsor Molecular Lab Procedures 2008

General rules:

1. Always clean up after yourself, including washing and putting away glassware and equipment.
2. You are responsible for replacing tips, microfuge tubes, etc. These supplies must be autoclaved.
3. Wear CLEAN gloves when touching any shared supplies such as pipette tips, microfuge tubes, etc.
4. If you spill anything on a tip box, microfuge tube plate, etc, sterilize with a 10% bleach solution and rinse thoroughly with regular water then ddwater.
5. All experimental materials should be clearly labeled with a project name, date, and your name when appropriate. If it is unclear what any stored samples are, we should be able to go back to the labeled date in your notebook and figure it out.

Autoclaving:

The autoclaves are located on the 3rd floor of Langley Hall.

1. Never completely seal jar lids. Put the lid on loosely, otherwise the change in air pressure can cause the jar to implode.
2. Put the items to be autoclaved into an empty autoclave and close the door.
3. Set the autoclave to the "wrapped" setting for 40 min of exposure and 40 min of exhaust.
4. Press start. If autoclaving doesn't begin within one minute, make sure the door is pushed all the way closed.

Dishwashing: Rinse thoroughly with warm water and scrub with a bottle brush. Use detergent only if something is particularly stuck on and then make sure to rinse thoroughly. Give all glassware a final rinse in ddwater.

Filling microfuge tubes:

Fill a clean beaker with tubes, cover with foil, and put autoclave tape on the foil. Autoclave.

Filling pipette tips:

1. Wipe down the area you will use with ethanol.
2. Use clean gloves to fill tips.
3. Place autoclave tape over the opening of the box,
   Autoclave

Micropipettor use:

1. Use only micropipettors you are permitted to use.
2. Never touch the tip end of the barrel.
3. Hold the pipettor vertically while pipetting.

4. Pipette up and down slowly, especially with the 100-1000 blue tip pipettor. You can easily suck liquid into the barrel when pipetting quickly.
5. Eject the used tip before setting the pipettor down. If you set the pipettor down with a little liquid in the tip, it can be sucked into the barrel.
6. Be VERY careful when mixing with a pipettor, and avoid mixing via pipetting if you are coming close to filling the tip (i.e pipetting 9 – 10ul with the 1-10ul pipettor, or 180-200ul with the 200ul pipettor). Never mix your samples with the 100-100ul blue tip pipettor.
7. Handle the pipettors gingerly. They can break or become uncalibrated by shaking, dropping, etc.
8. You can check calibration of the pipettors by pipetting a known volume of ddwater and weighing it. Water weighs 1g/mL so 1 uL should weigh 0.001 g, and 10uL should weigh 0.010 g, etc.

Refilling ddwater:

"dd" means double deionized. Obtain this water from the water filter in 157 Crawford Hall. To turn the water filter on, press the on/standby/setpoint button until the red bars are all lit up to the 12 mark. Then use the gun to dispense water. Put the filter back on standby before leaving, by pressing the on/standby/setpoint button until the red bar is rotating around the number 9. Never press the off button. Filters should be removed before turning the machine off so that ions/sediment don't collect in them.

If filling a carboy, turn the nozzle so that it doesn't lay in the bottom of the sink before placing the carboy in the sink.

Using the pH meter:

The probe should be filled with KCl silver saturated solution. Keep the probe wrapped in parafilm at the top, where it is filled, because the seal is broken and the liquid will evaporate without the parafilm. Store the probe tip in pH 4 buffer. Never store the probe in deionized water.

Calibrate each time you use the pH meter. Follow the instructions in the manual.

Rinse the probe in ddwater before using, and wipe off the water with a kimwipe. Do this between samples also. Then rinse in ddwater before storing in pH 4 buffer.