## Single emulsion particles using the homogenizer

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## Day 0

- If endotoxin free then soak in sodium hydroxide ON
- Make PBS by dissolving 1 tablet/200 mL DI water
- Make sure 3% (wt/wt) poly vinyl alcohol (PVA) (in PBS) is made. PBS tablets are used to create sterile PBS. For every 100 mL PBS, there will be 3 grams of PVA. Use magnetic stir bar and hot oil bath to quickly dissolve PVA. Otherwise place on hot plate stirrer overnight.

## Day 1

- 1. Weigh out 100mg of polymer in the small scintillation vials (white cap, silicon septa. Not the tall ones used for polymer synthesis)
- 2. Make sure that your drug of interest is weighed out in a small scintillation vial. Create a stock solution of drug such that for every 2mL of solvent, you will have the required amount of drug for 100mg of polymer. (solvent is variable for each drug).
- 3. Add the 2 mL of ethyl acetate/drug combination to dissolve the polymer.
- 4. While polymer is dissolving, place 100mL beaker with large magnetic stir bar onto stir plate. Create 0.3% PVA (in PBS) by adding 18 mL PBS and 2 mL 3% PVA (20 mL total is also variable)
- 5. Once fully dissolved, transfer polymer/solvent solution to a 50 mL falcon tube.
- 6. Add 12 mL of 3% PVA (in PBS) to the 50 mL tube to create the emulsion and homogenize for 30 seconds.
- 7. Draw up some of the 0.3% PVA from the stirring beaker and pour the contents of the 50 mL tube into the stirring beaker. Add the 0.3% PVA into the 50 mL tube to assist in gathering all of the particles.
- 8. Before using the homogenizer again, you must clean it. Fill a plastic reusable round bottom tube (above Mikes bench) with MilliQ water. Wash by turning on homogenizer briefly. Dump out the foamy water into the sink and fill again. Perform this 3 times. Rinse the homogenizer with ethanol or acetone. Wash 3 more times with MilliQ water.
- 9. Let the solution stir for at least 2 hours to allow for particles to harden.
- 10. After stirring, add particles to a fresh 50 mL conical tube and centrifuge for 10 minutes at 14,500 RPM at 4 degrees C to pellet (be sure to tare the tube if particle yield is important).
- 11. Decant and resuspend particles in 10 mL of water.
- 12. Repeat for 3 spins total (2 wash steps).
- 13. Resuspend particles in 5 mL of water (make sure completely suspended) and freeze particles.
- 14. Once completely frozen, place particles on the lyophilizer. Lyophilizer time is variable. When glass jar or 50 mL tube are no longer cold, particles can be taken off lyophilizer.