

Lindberg Lab Tips 9/09

- **There are no stupid questions.** Always ask about everything you are unsure of (and please, ask the right person – the one who is likely to know the most about that particular type of experiment! The PI can tell you this.)
- **DATE EVERYTHING YOU STORE with your initials and the date!!!** Everything you put in the freezers or refrigerators. The first sample in any long series bears the date and any important exptl. details (don't forget the year!) If your samples are important, CAP THEM. Label the SIDES of the white cardboard boxes, not just the tops. Try not to use anything but white cardboard boxes to store, and use as few as possible as we are running out of storage space.
- **BACK UP everything!** (especially cells; freeze them down before doing any experiments, and make sure you put them in liquid nitrogen within a week of freezing.) **Back up your computer** at least once a week on your provided external hard drive.
- **It is a good idea to leave 10% of your sample behind and freeze -so you can repeat if sample is off the standard curve.** The next day, if the experiment is successful, THROW THESE OUT.
- **ALWAYS FOLLOW SUCCESSFUL LAB PROTOCOLS** – small changes can easily add up to a total lack of success. And please don't change vendors without PI approval.
- **Make a cell entry in the card catalog and gray ampule storage book (both!) when freezing and also removing cells.** Do NOT take the last two ampules of any cell line without PI permission! (you must physically check that there are more- don't believe the records!) **Cross off any ampule you take with your initials and the date in the gray ampule storage book.**
- **When you get a new reagent in, make a card for the card catalog** and note the source AND DATE. Store it properly (usually in a Drierite box). Also, please write the month and year on the bottle when it comes in.
- **Don't usually put away stuff for other people when it comes in.** Put the package on their bench. If they are out for the day, then you may put it away at the approved storage temperature, immediately writing a note for them saying where it is.
- **Always transfer ALL of the information from any source reagent or vial you are given to your notebook.** Later, you will not remember what your source was (and neither will anyone else).
- **File a plasmid map** in the Blue Book within 2 weeks of new construction. Make sure the **location** of your plasmid is accurately specified (ie not just “in the freezer” but exactly where in the freezer). Label tubes with **backbone** and **species** and **antibiotic selection** as well as **insert** (“mPOMC-pcDNA3.1-Hygro”) and date and your initials. Do not use peel-off labels because they come off. Do not use any label at all for liquid nitrogen- just write on the tube. Use screw-cap tubes for N2, not Eppendorf tubes.
- **Thaw samples completely and mix gently before pipetting by inversion or pipetting. Vortexing denatures proteins. Solutions do not freeze evenly- so water is always on top.**
- **Understand the biochemistry behind every kit and tablet you use.** It is amazingly important.
- **Samples and standards must always be in the same buffer.** This is even more important.
- **Don't work alone on new things you might need help with- ie at night or on weekends.**
- Add 0.1% sodium azide to most solutions (but not solutions that ever touch animals or cells). It doesn't affect 99% of our enzymes at all -but mold will! Store all chromatography columns in azide solutions.
- **When working with a new protocol, read to the bottom before starting!** Make sure you interrupt at approved places if you stop in the middle (**ask!**). If you have an (approved) deviation from your written protocol on the day of the experiment, write changes down on the same sheet as the protocol- you never know when you will want to repeat the deviation -it may work better!
- Dilute your unknowns so that they fall in the **middle** of the standard curve (or repeat the assay to get them there).
- Don't accept replicate variation of greater than 15%. Something is wrong! You can do better! Redesign pipetting amounts- usually greater amounts will be more accurate. Less than 5 ul is very hard to do reproducibly. Using fine tips help.

➤ **Always do positive and negative controls to make sure your assay is working and is well-controlled.**

➤ Use different standard colors for control tubes and experimental tubes- I always label control tubes black, and exptl. tubes red.

➤ Put a standard lane on the left side of every gel- this way you will never have a direction problem.

➤ Put the handle of the Eppendorf tube on the outside when centrifuging; the pellet will then always be right below the handle.

➤ **Guanidine-HCl is VERY corrosive.** Please wash off everything that it touches well with distilled water.

➤ **Make sure you know how to seat a rotor,** the allowed speeds of each rotor, and the allowed speeds of each type of plasticware. If not you will lose your sample at best -and the (very expensive) rotor and centrifuge at worst.

➤ **Polystyrene (hard clear plastic) is very protein-adsorptive. Don't use it with dilute protein solutions in the absence of carrier protein.** Polypropylene is less absorptive, but many of our sticky proteins will still need detergent or carrier protein --or you will see time-dependent losses. (Can you tell polypropylene from polystyrene and polycarbonate?)

➤ **PLAN AHEAD!** Always do **feasibility calculations** the day before your experiment; also, make and chill your buffers the day before. Write out (typing is best) your protocols- the day before the experiment.

➤ **Be a good lab citizen-** clean an area up every few weeks that is not specifically yours, and if you use the last of something (or take the spare box off the shelf), order more the same day. If you do tissue culture, autoclave waste regularly!

➤ **Never throw out any primary data- always file it with the experiment.**

➤ **Do not keep two sets of records-** write everything you weigh on the same piece of paper that you put in your notebook. **DATE EVERYTHING- gels, blots, protocols- IMMEDIATELY.** File your results promptly in your notebook by PROJECT and DATE, not date alone.

➤ **Always write the PURPOSE of the experiment in a sentence at the top of the page!**

➤ **Always write the overall RESULT/CONCLUSIONS of an experiment on the first page so you can easily see what happened --years later. This need only be a sentence!**

This list has been made from accumulated errors over 25 years of running a laboratory.