

## Anseth Lab Orientation Checklist

In order for the lab to operate in the most efficient and safe manner possible, it is imperative that all lab members are familiar with lab policies and procedures. This checklist offers a description of essential guidelines for working in the Anseth Group.

### Safety

Lab Attire – Safety glasses and closed toed shoes must be worn at all times.

Material Safety Data Sheets (MSDS) – Detailed information about the reactivity, dangers, and handling of each chemical in the lab is located in alphabetical order in the MSDS Notebooks, which can be found on the counter in room 195.

Showers and eye-wash stations – 4 showers and 4 eye-wash stations are located in the lab. Be sure to familiarize yourself with their locations.

Fire extinguishers – 4 fire extinguishers are located in the lab. Be sure to familiarize yourself with their locations.

Fire Alarms – The nearest fire alarm is located outside of the lab in the hallway across from ECCH 199.

First Aid Kits – First aid kits are mounted on the wall and located in ECCH 191 (Hedco) to the left of the wooden chemical cabinet and in ECCH 192 and 159 near eyewash fountains. If you have a minor cut, please use these kits. If the injury is more severe (more than a band-aid), seek medical attention at the nearest medical facility and report the incident to IBC and EH&S.

Spill Kits – Whenever a volatile solvent is spilled, the area should be cleared and the spill should be soaked up with vermiculite (brown flaky stuff) stored in large cardboard box in back left corner of ECCH 199 (next to gas cylinders). Once solvent is soaked up by vermiculite, the solvent-containing vermiculite should be put in a sealable container, put in the hood, and sent away as solid waste.

Emergency Action Plan (EAP) – Refer to the established EAP. A quick reference is posted on each door of the lab.

Contact Numbers – Important phone numbers are posted on the lab doors.

EH&S Training – All members of the lab must complete the EH&S safety training prior to working in the lab. See the lab Safety Coordinators for details on how to complete this requirement. Training must be updated yearly.

## Lab Supplies

Glassware – There are two cabinets of assorted glassware which are distinguished by how they are cleaned:

*Brown Cabinet* (general use) – washed with soap and rinsed with DI water;

*Black Cabinet* (synthesis applications) – washed with soap and water, then put through successive base and acid baths before being rinsed with DI water.

Dirty glassware should be rinsed and put in either the tub by the sink in room 195 (for general use glassware) or in the sink in room 199 (for synthesis applications glassware).

General-use disposable supplies – Stocks of tips, tubes, pipets, gloves, and other disposable lab supplies are maintained for general use. These can generally be found on the shelves and in drawers around the lab.

*Overstock boxes* – most of these lab supplies have full cases that serve as overstock and are located on the top shelves above their lab storage location. When breaking into an overstock box, unpack the whole box so that it will be clear to the person that is doing the ordering that the overstock has been used.

*Ordering general-use disposable supplies* – In order to avoid duplication of orders, these general lab supplies are maintained by only one person in the lab. If you notice that one of these stocks is getting low, it would be helpful for you to write the item on the white board by Pete's bench. This is particularly true if you break into the last overstock box of an item.

Chemicals – Chemicals are stored in several locations, depending on their storage conditions and usage:

*191 Chemicals Cabinet* – General-use chemicals, stored in alphabetical order by name. Typically only solid chemicals are stored here.

*191 Chemicals Refrigerator* – General-use chemicals that require storage at 4°C, stored in alphabetical order by name.

*191 Flammables Cabinet* – General-use flammable liquids (organic solvents, alcohols, etc.).

*191 and 192 Acid Cabinets* – General-use acids are stored under the 191 and 192 chemical hood.

*191 Base Cabinet* – General-use bases are stored in the cabinet by the sink in 191.

*PEGs* – General-use PEGs are located on the shelves above the weigh bench in 191.

*Equipment reagents* – Specific chemicals are required for the operation of certain instruments in the lab. Often, these stocks are kept in the area where they are used. Do not use these stocks for other purposes unless cleared by the person who is responsible for maintaining the instrument.

*Personal chemicals* – Each researcher in the lab may have reagents that they have ordered specifically for their project. These chemicals are generally stored at that person's bench or in their designated storage areas in the lab refrigerators and freezers. Before using these personal chemicals stocks, it is imperative that permission is granted from the lab member to whom it belongs.

**Ordering chemicals:** If you empty a general-use chemical, it is your responsibility to make sure that a replacement has been ordered. If it has not been ordered (or is not already in stock), put the chemical in the ordering log. If you notice that a general-use chemical is getting low, please put it in the ordering log so that it will arrive before the old one runs out.

## Waste Disposal

Glass Containers – Anything that has the potential of being sharp, but is not a syringe, needle, or knife. These regular sharps include non-biologically contaminated:

1. *pipet tips*
2. *plastic pipets*
3. *glass pipets*
4. *broken glass*
5. *microscope slides and coverslips*

Note: Pipet tips can be collected in a secondary puncture resistant container. When full please seal the container closed and label as non-hazardous waste.

Sharps Waste Containers –The following items are NOT considered “regular” sharps and must be disposed of by placing them in specific, puncture-proof waste containers (large red plastic containers labeled “Regulated Medical Waste”):

1. *Needles*
2. *Syringes*
3. *Razor blades*
4. *Scalpels*
5. *Scalpel blades*

Syringes are considered “sharps” because of the public opinion that they are biohazards. Therefore, they cannot be sent to the landfills and must be handled as hazardous waste.

Sharps that are used for biological applications must also be autoclaved. Biohazardous sharps containers are located under the hoods in each tissue culture room.

Biohazardous waste – All materials involved in cell culture (bacterial or mammalian) such as media bottles, pipet tips used for tissue culture, agar plates, etc. Basically, if there is a chance that something will be able to grow on it, it needs to be rendered not biohazardous via autoclaving or bleaching.

*Syringes, needles, glass pipets and pipet tips* – All go in the red biohazardous sharps containers labeled “Regulated Medical Waste”.

*Media bottles, plastic pipets, plastic plates* – All go in the containers lined with red biohazard bags.

Paper/Cardboard – Bins are located around the lab for recyclable paper.

Regular trash – Anything that is neither recyclable nor potentially hazardous (package wrapping, tip containers, non-contaminated paper towels etc.).

Chemical waste – Chemical wastes should be separated based on the type of chemical present: (1) organic solvents, (2) acids, (3) bases, and (4) organic and aqueous wastes. Here are a few general rules.

*Organic solvents* – Organic solvents only (no contaminants, reactants, or byproducts from reactions) should go in the 20 L plastic carboy under the hood in ECCH 195. Document the volume of solvent added on the sheet with the carboy. This carboy should be vented (back valve should have hole in it). **DO NOT ADD REACTION WASTE TO THIS CARBOY; IT CAN REACT AND CAUSE EXPLOSION.**

*Acids* – Acids should go in the 20 L plastic carboy under the hood in ECCH 195. Document the grams of acid added on the sheet with the carboy (note calculation on how to do this on sheet). This carboy should be vented (back valve should have hole in it).

*Bases* – Bases should go in the 20 L plastic carboy across from the hood in ECCH 195. Document the grams of base added on the sheet with the carboy (note calculation on how to do this on sheet). This carboy should be vented (back valve should have hole in it).

*Mixtures* – Reaction waste containing various solvents, reactants, catalysts, and other types of organic compounds should be collected into a personal waste container that you are responsible for maintaining this vessel, documenting the chemicals within it, and submitting a waste tag to EH&S when the container is full.

- You should separate the chemicals into different waste containers so that you do not mix molecules that can react with each other (MIXING CHEMICALS THAT ARE REACTIVE CAN LEAD TO EXPLOSIONS WITH FLYING GLASS SHARDS AND LAB FIRES!).
- Specifically any violently reactive molecules should be quenched prior to disposal; seek assistance if you do not know how to do this.
- When you are adding chemicals to a waste bottle, do the addition in the hood with the sash lowered.
- Leave the cap on the bottle loose to allow for vapor expansion with temperature changes.
- Store the bottle in an SAA when not adding chemicals to it.
- Do not fill a bottle more than 3/4ths full (head space is need for vapor expansion, otherwise bottles can explode!

Empty Chemical Containers – When you use the last of a chemical reagent, clean the empty container and place it on the cart beside the autoclave for disposal. Depending on the reagent, the protocol for cleaning the container will be based on the nature of the chemical. If you are not sure how to properly clean the container, ask someone who knows.

Unknown – If you are unsure of how to properly dispose of something, you must ask for help. Improper waste disposal can lead to dangerous situations, environmental contamination, large fines, etc.

## **Equipment Usage**

The equipment in this lab is extremely expensive and must be used with care. Misuse can result in costly repairs that hinder the success of the lab and its members. Additionally, improper use of the equipment can result in dangerous situations that are potentially life-threatening.

Equipment Protocols – Extensive protocols have been written for each of the major instruments in this lab. These protocols should be used to train new users and as a readily-available reference. Written protocols DO NOT substitute for hands-on training obtained from the person(s) responsible for the equipment.

Sign-up sheets – Many of the instruments are in high demand and an effort must be made to accommodate multiple users. Sign-up sheets are posted by each of these machines. Use them to block out times that you know you will use them. Be considerate. That is, don't sign up for the entire day when you only need it for a few hours. Also, update the list if your plans change and you know that the machine will be free during a time that you had signed-up to use it.

Undergraduates (UGs) – Due to the nature of specific equipment in the lab, undergraduates are not permitted to use the following instruments without direct supervision: confocal and new time-capture microscopes, Laser Capture Microdissector (LCM), peptide synthesizer (SPPS), HPLC, and GPC. For all other equipment, it will be the mentor's responsibility to properly train and oversee UG equipment usage. UG mentors will be held responsible for improper use of lab equipment by their UGs.

When in doubt about how to use any piece of equipment in this lab, you must ask for help. Failure to use equipment properly and by the guidelines described above will be viewed as a breach of lab policy.

## Ordering

Ordering is done by placing the item to be ordered into the “Anseth Ordering Log” spreadsheet that is located on the TC1 computer. Only graduate students and post-docs are authorized to place orders.

Chemicals – Please write out the full chemical name and include the lab storage location. Upon arrival, the chemical must be entered in the inventory and assigned a bar code. Leave all chemicals to be entered into the inventory in room 159 on the counter next to the sink.

Orders over \$500 must be pre-approved by Kristi.

Orders must reach a \$50 minimum for each vendor before they can be placed by Cathy. If an item is less than \$50, it will be ordered only when other items from the same vendor makes the total greater than \$50.

## Lab Resources

Chemical Inventory – The undergraduate computer contains an inventory of all of the chemicals that are currently maintained in the lab, where they are stored and an electronic copy of the MSDS. This is a useful resource for finding reagents. Also, check this list prior to ordering reagents, as the item you want may already be stored in the lab.

Anseth Group Protocols – Useful protocols have been written for commonly used techniques and can be found on the group website:  
(<http://www.colorado.edu/che/ansethgroup/index.html>).

Molecular Cloning by Maniatis – Considered the “bible” for molecular biologists, this 3-volume set includes detailed protocols, as well as, technique theory for most biologically-based assays. These volumes can be found on the bookshelf in room 195.

Vendor Catalogues – An assortment of frequently used vendor catalogues can be found on the bookshelf in room 195.

Anseth Group Plasmid Log – A log of the DNA plasmids that are currently stored in the lab is located on the bookshelf in room 195. The log contains plasmid maps as well as the storage location. If you would like to have one of these plasmids, ask Pete about how to go about getting it.

NMR Spectra – This 3-volume set gives NMR spectra of many common compounds. These volumes can be found on the bookshelf in room 195.

**By signing below, you acknowledge that you have read the information contained in this Anseth Lab Orientation Checklist and agree to abide by the policies outlined therein.**

**Name:** \_\_\_\_\_ **Signature:** \_\_\_\_\_ **Date:** \_\_\_\_\_

**By signing below, you acknowledge that you are a qualified member of the Anseth Group and that you have personally gone over the information contained in this Anseth Lab Orientation Checklist with the person named above.**

**Name:** \_\_\_\_\_ **Signature:** \_\_\_\_\_ **Date:** \_\_\_\_\_