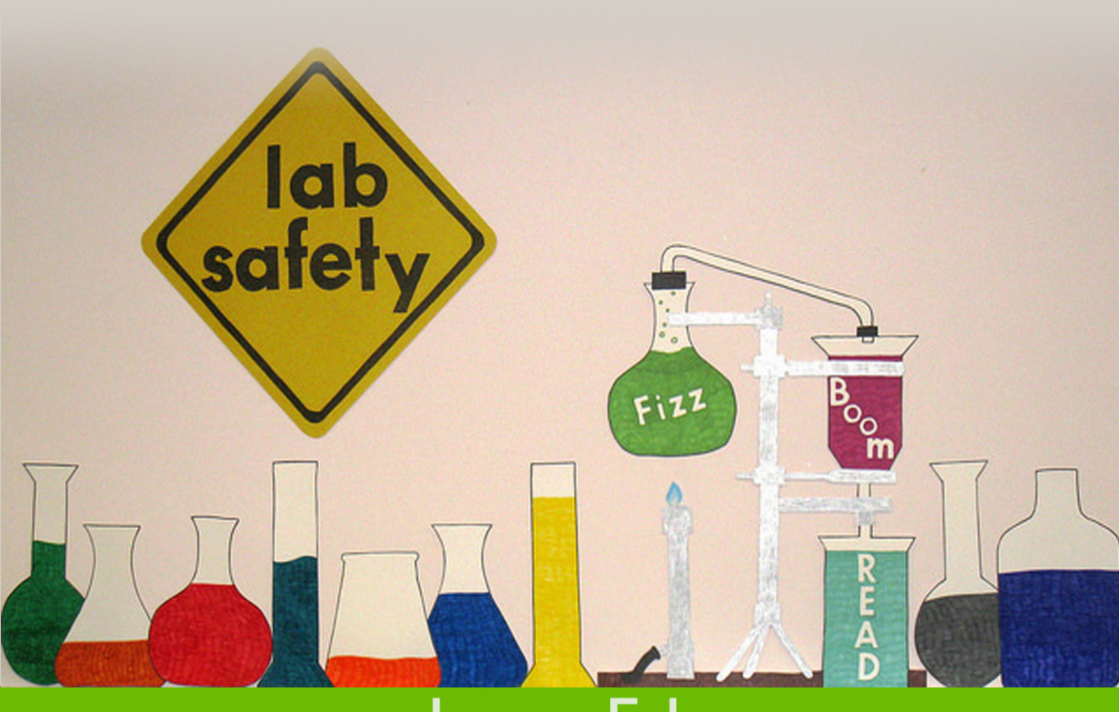
The BitesizeBio Guide to Lab Safety



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Chapter 1
Introduction:
The basics of lab safety

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When you've been given the task to run a new experiment, it can feel like you've been left to "sink or swim" because of the assumption that you have 100% of the skills and knowledge required to complete it safely. But no one should ever how to wonder about how to perform lab work safely!

The purpose of this ebook is to introduce you to some of the most common principles of scientist's safety topics that you will encounter every time you step in a laboratory. On top of that, it will help point you towards additional resources to answer your questions when deeper questions about the topic arise.

At first glance, the lab can appear to be a very dangerous environment. However, much like everyday household items, lab tools, reagents and equipment come with a user's manual, so you can learn how to work with them confidently. With proper knowledge and practice, your lab will become a very safe place to work!

While this ebook assumes that you have some prior exposure to chemistry, biology and general laboratory techniques, fear not if you are new the field! Let this brief guide serve as a companion at the bench; it describes some of the typical environments you'll encounter, including common pitfalls, dangerous scenarios and how to better avoid and prevent them.

Chapter 2
Personal protective equipment: Dress for success

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Personal protective equipment (PPE) comes in all shapes and sizes. Wearing the right PPE is a minimal expense – both in terms of your time and the lab's money – compared to the cost of injury or death that could occur if PPE is never used at all.

It's a good idea to take stock of your lab's inventory to see what is available now, what fits (and what doesn't) and what is best for you and the experiments you're performing. What's right for your colleagues and collaborators may not be right for you, so talk with your lab manager about keeping stocks of the right PPE that you need.

Lab coats

Lab coats are the simplest and most basic form of PPE. Lab coats are typically made out of thick white cotton, fall to the knee and have long sleeves. They're designed to provide a basic layer of protection against heat and cold, splashes, spills, and stains (for instance from bleach). A lab coat is reusable, so the better care you take of it, the longer it can be used as mid- to upper-body protection.

How to wear a lab coat

When it comes to lab coats, sizing matters! The ideal lab coat should fit relatively close to your torso, should button without straining and should have sleeves that end at your wrist. Sometimes you have to work with what's available in the lab, and that means the fit may a bit too small or a bit too large. On larger coats, you can just roll up the sleeves to reach your wrist. Though be mindful of contamination: rolling a sleeve *in* potentially exposes you to anything that was left over on the coat. Rolling *up* exposes the inside of the coat during an experiment, which means the next person might get an unwelcome surprise when they wear it. To get the most protection out of your PPE, wear it the way it was designed to be worn. Tie it, button it, or zip it all the way to cover you completely.

How to take care of a lab coat

There's nothing quite like having your own lab coat – or two: one for primary use, the second to use as backup when the first is being washed. It's *your* lab coat, so you know exactly where it has been, where it stays, and how often it gets used.

FIGURE 1: How to hang up a lab coat. Source: Vivien Rolfe. Licenced under CC-BY-SA-2.0.

If any lab coat looks like it was used to mop the floor, that should give you pause. You don't want to have to choose between wearing a lab coat with mystery spots and taking your chances with known hazards in only your street clothes. Play it safe by washing your lab coat regularly, especially when you know that it has become soiled. Coats don't provide invulnerability either, so if you suffer a splash, take it off and have it washed so that it can live to see another day.

Clip any stray threads so that there is zero chance that any part of your lab coat will drape into or through your experiment. The same goes for loose buttons: sew them up so they don't catch on anything when moving around lab equipment!

When you're not using your lab coat, hang it in a designated spot that is separate from where you hang your everyday coat (to avoid transferring any contamination onto your "normal" clothes). Each lab coat should have its own hook: one lab coat hung on top of another transfers contamination from the outside of one coat to the inside of another, which ultimately ends up on the clothes you wear (*Figure 1*).

When not to wear a lab coat

It seems to contradict common sense, but scientists have been known to wear their lab coats in public spaces, like the cafeteria. You wouldn't think about eating while in the lab, so why would you wear your lab coat while eating outside of the lab? You risk ingesting harmful chemicals, wiping your face with a dirty sleeve, touching surfaces that others touch after you and spreading lab bugs everywhere. Here's where you have the power to help protect others, by wearing a lab coat in the right place and the right time: in the lab when actively running experiments.

The hard and fast rule about lab coats – as is with most PPE – is that they should only be used in the lab!

Gloves

After lab coats, gloves are the next basic element of PPE. At about 10 cents per hand, they're also probably the most inexpensive shield you'll ever wear. Make a conscious decision to wear them religiously, but take care not to sabotage that protection by using them the wrong way!

How to use gloves effectively

Gloves protect you from the outside, inward, so don't drain that protection from the inside out. Rings with a high setting, prong or pointy edge can rip through the material like a hot knife through butter, exposing bare skin to dangerous reagents, just as if you were wearing no gloves at all!

Another way to weaken the shield that gloves provide is to slather lotions, hand sanitizers and other creams on your skin before donning a glove. Many consumer products melt the protection away in the same manner as if you exposed the glove to a strong chemical reagent. Clean, dry hands make gloves easier to wear, which is the best way to protect your skin.

Consider using double gloves and nitrile gloves for specific applications. Nitrile gloves are the popular choice for lab PPE because they're as close to "all-purpose" as you can get. The material shields you from a wide range of reagents while providing excellent range of motion and adequate protection against pokes, too! Although you give up a little bit of your range of motion for double the protection, two pairs of gloves strengthens your defenses. The stronger your shield, the more difficult it is to pierce.

In addition to latex and nitrile gloves, most labs have one pair of thick orange gloves for use with the autoclave and thick blue gloves for use with anything colder than a regular freezer. These aren't disposable, so we'll talk more about them later (see *Extreme temperatures: autoclaves, freezers and liquid nitrogen*).

Different experiments require different types of gloves. Ask your lab manager about a glove reference guide, especially if an experiment calls for chemicals more exotic than what you use everyday.

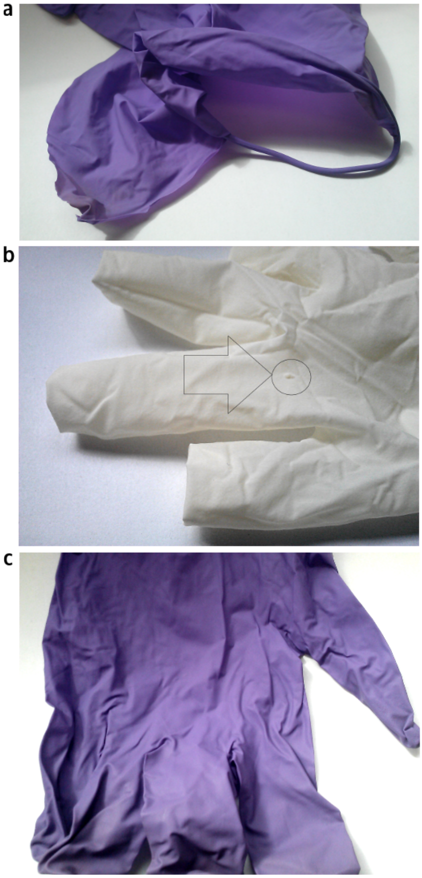


FIGURE 2: Faulty gloves: a) torn, b) blemished and c) brittle. Source: Jason Erk.

How to store gloves

To make sure that gloves last up to (and hopefully beyond) the expiration date, it helps to keep them stored in an area that is away from direct sunlight, heat and ultraviolet light (such as the cell culture hood and gel imager), and in a room with a constant climate.

Before you put a glove on, take a quick look to see if there are any small holes or if the color just doesn't seem right. When you put the glove on, stretch it gently to identify weak areas that may rip or tear (*Figure 2*). If you are uncertain, discard the glove and try another one. If your glove has torn once, it's a telltale sign that there may be other weak spots that you can't see yet. To be safe, wear a new glove (or double up) before you find out that the glove you have on is ready to rip further.

When to change your gloves

The longer you use a glove, the more items you touch – and that means the potential for contaminating experiments, tools and possibly even you. You know what to do when a glove *is* contaminated, but if you don't realize that it is, you may be unnecessarily exposing yourself in the long run. That's why it's a good idea to keep tabs on how long you've worn a pair of gloves.

Gloves don't confer invulnerability, either. The longer they are exposed to a contaminant, the greater the chance that it's soaking through the glove, which means that in time you, too, will be exposed. If your gloves become soiled, take them off and wash your hands as soon as you can.

When removing your gloves, keep the contaminated surface away from your skin. De-gloving made simple: gently pinch along the wrist of one glove and pull it off the hand (without tearing it!). Then carefully reach inside the cuff of the other gloved hand, along your wrist to pull it off, turning it inside out over the first. If there's any contamination on the surface of either glove, you've just avoided touching it completely! If you're curious to see how this is accomplished, check out this great video tutorial.

Some people will remove a glove and try to reuse it later by blowing it up so their hand can slide in a second time. This practice not only contaminates your experiment, but you too, as the dirty used glove is brought in direct proximity to your mouth, nose and eyes! When you remove a disposable glove, discard it. Then grab a fresh pair to protect yourself.

There are a few common reasons why someone would think about not regularly changing their gloves. First, it saves the lab money, even if it *is* only a handful of pennies. Also, gloving and de-gloving often is uncomfortable because of sweaty hands, difficulty in putting on a fresh pair or multiple uses of powdered gloves drying your skin out quickly. Unfortunately, these practices put everyone at risk for exposure. So protect yourself and your colleagues by practicing good glove hygiene.

When not to use gloves

If you take a moment to pay close attention to what you touch and where your hands are, you may be surprised to discover all the things that you want to (and maybe even do) touch while wearing gloves, without even thinking about it.

The hard and fast rule for *not* wearing gloves is this: if you're about to do an everyday task that you regularly do outside the lab, remove your gloves first and wash your hands. That way you're not spreading contamination around.

Here are some real-life examples of where you should not wear gloves:

- **Adjusting eyewear, rubbing your eyes, etc.** I know it's tempting to nudge your glasses back up your nose, but this brings your potentially contaminated hand very close to your face and eyes. Make sure you're comfortable and prepared to work before putting on your gloves to avoid this situation.
- **At a computer.** Computer keys are a shared surface. If they're contaminated, it's a very easy way to spread it all around the lab to door handles, clothes, hair and more.
- **At your desk.** It may look like a hurricane hit, but it shouldn't be a biohazard zone. Contamination away from the bench is harder to keep track of because you risk taking it home with you.
- **In the lab's attached break room.** Peanut butter and *E. coli* sandwich? No thank you!

- **In common areas, like in the hallway and when opening doors.** People in public spaces may not realize that you just put on a clean pair of gloves. To them, it looks like you are in the middle of an experiment, and are bringing whatever bugs are in the lab along with you. Wait to wear your PPE until you are at the bench. If you must wear gloves outside of the lab, wear one glove on one hand only (see *Moving/transporting reagents*) so you are able to open doors without causing undue worry.

When you share equipment, supplies and bench space it is beneficial to have a lab policy on when and where to wear gloves. An easy way to accomplish this is to place keywords and phrases in strategic areas of the lab. We've used "GLOVES REQUIRED" and "NO GLOVES" signs to denote where new PPE should be worn (click here to access printable versions). It's a small effort, but pays big dividends in terms of encouraging others to consistently wear their PPE.

Face and head protection

While face and head protection are not required for every experiment, they are important to wear during certain applications to shield crucial areas where chemicals can enter the body (like the eyes, ears, nose and mouth). In this section, we'll talk about how to choose and use face and head protection.

Choosing the right eyewear

The simplest form of eye protection is safety glasses (*Figure 3*). Easy on, easy off, they repel flying objects and small splashes. Splashes are a big concern when working in any lab, even when doing a basic task like washing glassware, so opt for greater safety by selecting goggles that form a protective seal around the eyes (*Figure 3*).

a

b

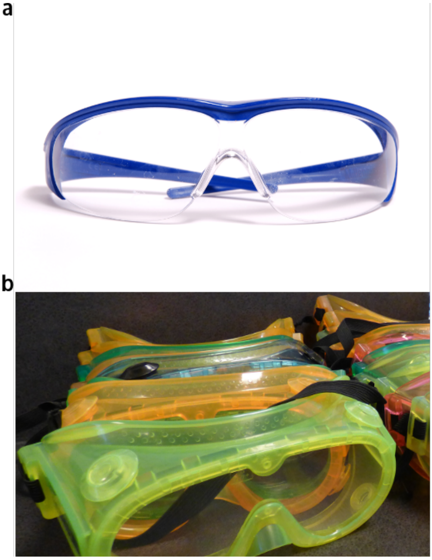


FIGURE 3: (a) Safety glasses and (b) splash-resistant safety goggles. Source (a) Lilly_M . Licenced under CC-BY-SA-3.0 (b): Jason Erk.

Adjust the strap to get a comfortable fit. Too tight and you'll know by the heavy goggle outline around the eyes when they're removed. Too loose and they'll only protect your neck! Most goggles will even fit over prescription glasses, so you can get a double layer of protection.

Protective eyewear is a very personal option. When you have your own pair, it is adjusted to fit you and lives nearby so you always know where to find it. It's great if your lab has shared eyewear protection available, but like lab coats you won't always know how much use and abuse the goggles have been through.

One situation in which all scientists use eye protection is when working in areas with ultraviolet (UV) lights – although you need to make sure you're using UV-rated safety goggles. Staring directly at a UV light source is like looking directly at the sun. UV-rated goggles are good for most general applications. Choose a UV-rated face shield when working with nucleic acids and UV illuminators to protect your face as well as your eyes.

FIGURE 4: a) Filter mask and b) N95 respirator (bottom). Source (a) Blossoma. Licenced under CC-BY-SA-3.0 (b) By Banej. Licenced under CC-BY-SA-3.0.

Masks

The laboratory environment can expose you to airborne powders, aerosols, animal dander, chemical fumes and gases, just to name a few. Covering your mouth and nose with a face mask filters the air you breathe, which helps protect your body from chemical exposure and chemical sensitization.

Thankfully, masks are easy to use. Disposable filter masks (*Figure 4*) are built with a metal strip that sits on the bridge of the nose – you can set the perfect fit in a pinch. Where there's a box of gloves for anyone to use, there is usually a box of filter masks nearby. Like gloves, these filter masks are one-time use only, so throw it away when you're done with your task.

Other, more specific environments may require a greater degree of air filtering, sometimes up to 100% of the air breathed. For these applications, you'll need to use an N95 or N100 (*Figure 4*) mask. You will find these masks especially helpful for any work that involves animals. Repeated exposure to dander and other allergens can produce the same response as if you were repeatedly exposed to a chemical reagent. Over time you can become sensitized, so that smaller and smaller amounts of the allergen will trigger an allergic response.

Hearing protection

Another type of PPE that may not get as much attention as it deserves is ear protection. If you use a sonicator to disrupt cells, work around large, noisy machinery or even have equipment that blasts loud, white noise, add ear plugs and noise-dampening headphones (*Figure 5*) to your arsenal of PPE.

FIGURE 5: Noise-dampening headphones. Source: AGoodGuinness Licenced under CC-BY-SA-3.0.

Chapter 3
Common lab hazards

Image source: CSIRO, licenced under CC BY 3.0.

Now that you know how to protect yourself when working in the lab environment, let's talk about the kinds of hazards that you may encounter on a daily basis. In general, these hazards will fall into one of the following categories: chemical, biological, radioactive and technical (i.e. lab equipment and tools).

Chemicals

Solid, liquid or gas: each chemical phase behaves differently, and it's up to you to avoid exposure to and sensitization from chemical reagents. In this section, we'll talk about how to handle chemicals safely.

MSDSs

Your first stop for information about any chemical should always be the Material Data Safety Sheet (MSDS). MSDSs are the instruction manuals for chemicals – what they're made of, how to handle them, how to dispose of them and more. Even the most seemingly innocuous, common reagent (water, salt or sugar anyone?) can have an MSDS that reads like a horror story. However, each MSDS has the same 16 categories so you can quickly learn the important facts about your reagent and handle it safely and confidently!

1. The **Identification** section describes your item (including common names and synonyms), the supplier or manufacturer and what number to contact in case of questions or emergency.

2. The **Hazards** Identification section is your "abstract" to the hazards of the chemical. Hazards are described by intensity on a scale of 0–4 (0 = least intense, 4 = most intense), and are often

depicted in a colored diamond or hazard information bar for quick examination. This is your key to identifying the immediate impact to health, flammability and reactivity; the specifics are discussed explicitly in following sections.

3. The **Composition & Ingredient Information** section describes the makeup of the reagent. Some MSDSs will list individual ingredients; all will include whether or not a particular ingredient is hazardous. Some MSDSs are more inclusive, listing individual chemical names, formulas and molecular weights in this section.

4. The **First Aid Measures** tell you what you need to know to render aid to your colleagues in the case of exposure. You will be a "first responder" in case of spillage or exposure until backup arrives.

5. **Firefighting Measures** are important to have on hand because not every chemical fire is fought with water. Some reagents decompose as they burn, creating secondary, equally intense hazards that you will need to account for.

6. The **Accidental Release Measures** section explains what you need to do to protect yourself and the environment in the case of a spill, and where possible, how to clean up (see Spills).

7. **Handling & Storage** is self-explanatory, though it notes any special circumstances, like if your chemical readily soaks up moisture (i.e. is hygroscopic), including what conditions to avoid (i.e. don't store acids and bases together).

8. The **Exposure Controls & Personal Protection** section describes the PPE (see *Chapter 2: Personal Protective Equipment*) required to protect your lungs, eyes, hands and body when

handling the chemical. It defines any exposure limits applicable to the ingredients in your reagent and helps you decide whether or not to request exposure monitoring.

9. The **Physical & Chemical Properties** section lists technical data, including molecular weight, color, odor, pH, boiling/freezing/melting points, flash point and vapor pressure, among other interesting information that can help you distinguish one chemical from another.

10. **Stability/Reactivity Information** identifies the conditions that will make your reagent an "angry" chemical, such as: shock, static electricity or ambient temperature. Here's where you can find out if a chemical reagent needs to be combined with an additive to maintain stability, and whether or not there are telltale signs (i.e. color changes) of spoilage to look out for.

11. **Toxicological Information** alerts you to the bad things a reagent can do to your body (target organs), whether it causes cancer (carcinogenicity), affects your unborn child (teratogenicity) or your DNA (mutagenicity), and includes the LD50.

12. The **Ecological Information** section alerts you to the bad things that a reagent can do to the environment, how it accumulates and how much it degrades (or doesn't), including the EC50.

13. **Disposal Information** describes not only how to discard your chemical when you're done using it, but how to discard any contaminated packaging.

14. **Transportation Information** lists the shipping requirements that apply when the item is shipped from manufacturer/supplier to you, or from you to someone outside of your organization.

15. **Regulatory Information** contains required notices at the national and state levels.

16. **Other Information** is the most fluid section in your MSDS. Some manufacturers add the date of document creation and updates here. Others use this section to grant you a right to print unlimited copies for internal use. Others state that the information given in the MSDS is current, but that its purpose is to be used as a guide.

Every time you order a new chemical, file the MSDS in your lab's collection (if it does not already have one). For the reagents you order regularly, think about making a schedule to check whether or not your lab has the latest, updated version. That way you know your records are complete and up to date. You may also want to consider building your own MSDS library for the chemicals that you use most often. If you keep electronic files, you can organize and name each MSDS by item name, manufacturer, assay purpose, etc. in a way that is useful to you.

Storing chemicals

Generally, different categories of chemicals are stored in separate cabinets based on temperature sensitivity, light sensitivity and reactivity. Two of note – acids and bases – are separated for good reason: remember that an acid/base reaction is much like your at-home baking soda and vinegar volcano (except on a more intense scale). Separately, the two are highly corrosive. Together, they neutralize one another, creating carbon dioxide gas and heat! That's why acids and bases should be stored in separate, dedicated cabinets. Another major category is anything that is flammable, volatile or with a high fire hazard rating...into the flammables cabinet it goes!

Many chemicals are stored on the shelf at room temperature, while others are stored in the refrigerator and just as many are stored in the freezer. Temperature is a major factor in whether or not a chemical remains stable over a long period of time. For other reagents, temperature can affect volatility. Take dry ice and liquid nitrogen as examples: one sublimates at room temperature, while the other boils violently. The effect is slowed when the two are kept cold.

Some reagents are light-sensitive and need to be stored appropriately to prevent degradation. For instance, both tetracycline and hydrogen peroxide undergo a chemical reaction when exposed to long bouts of ambient light, rendering them less effective. Light-sensitive chemicals often come in amber bottles, which will protect them from light when stored on the shelf. Another option is to wrap the tube or bottle with aluminum foil.

When storing chemicals, be sure to use a container that is chemically compatible. For example, are you working with chloroform? Double-check that a plastic waste container is strong enough for the task. Otherwise you may find a surprise...that the chloroform waste melted through your plastic container and all over the cabinet. Be aware of expiration dates and shelf life, even if they're not listed on a container. It is worth noting that over time, some chemical reagents can present new hazards that differ from when you received them. Take our friend chloroform again: repeated use of a bottle exposes it to oxygen, which encourages decomposition into phosgene. Another interesting chemical of note is 2-methylbutane (also called isopentane). You might use it to snap-freeze fresh tissues, but it has a relatively short shelf life of about a year.

Chemical storage conditions are printed right on the chemical label. You can also find them on the MSDS or product insert. How you take care of your chemicals from the day you receive them until the day you discard them ultimately determines whether or not they live an uneventful life in the lab.

Using a fume hood

The effect of brief chemical exposure can range from dizziness to irritation of the lungs; prolonged exposure can cause permanent harm that may not be apparent until later. So when you're about to begin an experiment that uses odorous or hazardous chemicals, choose to run it inside the fume hood instead of at the bench.

A fume hood is an enclosed space with negative air flow. What this means is that air is sucked out of the lab through the hood and vented outside, thus keeping any harmful fumes or substances out of the lab air environment.

Before you begin working in a fume hood, do a quick safety check to make sure that it's working properly. An active fume hood will have a pressure gauge that reads higher than zero. A basic way to check that the hood is operational, however, is to tape a lab wipe or paper towel to the lip underneath the sash. When you see it dance in the wind, that shows you that the air is moving through the hood!

Place your tools and equipment in the center of your workspace, set back from the front of the hood. Keeping chemicals away from the front of the hood ensures that any hazardous fumes are drawn up through the chimney, rather than finding a way to spill back into the lab.

Once you have all of your materials and equipment set up inside the hood, lower the sash so that the opening is high enough for only your hands and arms to go inside! Keeping the sash lowered helps direct air flow up the chimney and doubles as a splash guard when working with hot or boiling solutions.

Above everything else, keep your head outside of the fume hood! It seems intuitive, but you actually defeat the purpose of the hood when you place your head, directly over your materials. The lab will be protected, but you will breathe in everything that can harm your body.

When you've finished your experiment, you may want to leave the fan on for an extra minute or two to ensure that all of the fumes have been sucked out.

Moving/transporting reagents

Moving reagents within the lab environment can create new, potentially hazardous situations, so think ahead: being proactive helps you respond to any incident more quickly and more safely.

Keep the following tips in mind:

- **Carry only what you can hold comfortably** – don't overload. Carrying too much at one time is a sure-fire way to invite accidents. You may be tempted to save time by loading your arms with a precarious stack of boxes, bottles, or carboys – but play it safe and make a few trips instead.
- **Close tube and bottle tops securely** when not in use. Luck may smile on you if you accidentally tip over a container and it doesn't break, but a bottle with an open top is sure to spill. If the

top is closed when not in use, there will be no surprises when a lab mate goes to pick it up and carry it somewhere else in your lab.

- **Carry items upright and level.** It's good practice to always carry chemical bottles, boxes, etc. as if the top isn't on securely – because someday it might not be! Treat chemicals with respect by carrying them carefully and prudently.

Be particularly careful when transporting reagents between labs. This takes you in and through public space, which is a relatively uncontrolled environment. Plan to bring PPE with you, just in case. Here are a few good options:

- **Glove one hand**. For short distances, one glove is perfect to hold onto an item like a small bottle or conical tube after you have filled it from a stock bottle. You can then use your ungloved hand to touch door handles and elevator buttons, reassuring others in the public space that no reagents are about to come in contact with their hands.

- **Use a smaller, secondary container**. When working with chemicals that are stored in bulk quantities, a smaller container filled with a working volume of your reagent is much more manageable. If the bottle is dropped and shatters along the way, it's easier to clean up 40 ml than 4 L.

- **Use a cart and a bin**. When transporting chemicals for long distances, place your reagents in a bin and wheel them on a cart. This reduces any opportunities for slips, falls and dropped bottles. Alternatively, carry your small bottle inside of a protective box. That way you won't need to wear gloves in public!

- **Move new reagents in their original packaging**. Reagents shipped from the factory are packed with absorbent material in case of spillage. After you've checked it in with your lab manager, transport the reagent to its final destination in its shipping box and unpack it there!

Reagent and chemical disposal

Now for the question of the day: "Can my reagent be poured down the drain?"

To be safe, the general rule of thumb is that *no* reagent or experimental waste should ever be poured down the drain. Of course, there are exceptions to this rule. Look to your organization's laboratory safety documentation for two very important disposal lists. The first one is fairly short, describing what reagents may be permitted to be disposed of down the drain. Usually this will list a number of criteria that have to be met first, like near-to-neutral pH, diluted concentrations and non-hazardous liquids, just to name a few. The second list is the one to frame prominently in the lab: the "DO NOT DUMP" list. This list will alert you to which reagents need to be disposed of by your organization's chemical waste pickup program instead. The MSDS (see *MSDSs*) is an excellent source of information if you're not sure how to dispose of a particular reagent.

Take an active role in managing your lab's chemical waste by doing the following:

- **Label your waste containers**. Different types of experimental waste require separate waste containers. Remember how acids and bases are stored separately? Incompatible chemical wastes demand the same care when being held for disposal. Label waste containers with the contents and concentration to avoid accidentally mixing chemicals that may react poorly.

- **Have waste containers removed regularly**. It's hard to tell how much liquid waste is in an amber bottle – better be safe than sorry and have it emptied regularly. Besides, a stock of waste containers takes up valuable research space, especially if the carboy lives inside the fume hood. Check with your lab manager or safety department to schedule a waste pickup.

Biologicals

When you work in a biology lab, you may be asked to handle bacteria, tissue culture cells, viruses and other living organisms. To protect yourself, it's good practice to perform any manipulation within a biological safety cabinet or tissue culture hood.

Using a tissue culture hood

To keep your environment – and your cell's environment – as contaminant-free as can be, the tissue culture hood (also known as a "biological safety cabinet") provides two-fold protection by creating a force field of air that washes any airborne bugs towards a filter, and a glass window that is your shield against any splashes caused by vigorous pipetting (or a super-charged automatic pipettor!)

Using a tissue culture hood is quite simple when your lab has established a "how to guide" that essentially says "clean as you go in and clean as you go out." Here's a general idea of what you can do:

- Turn the UV light on ~10–15 minutes before starting your work (this time will vary from lab to lab) to sterilize the interior of the cabinet.
- Then turn off the UV light and turn on the fluorescent light. We wouldn't want the high-energy UV lamp to destroy our cells!
- Turn on the blower – this cleans the environment when you move items in and out of the hood.
- Adjust the windshield up a couple of inches. Hear the buzzer? Now it's open too far. That reduces the effectiveness of your protection.
- Wipe down the working surface with 10–30% bleach.
- Wipe down your working surface with 70% ethanol.
- "Spray in, spray out" – every working item that goes into and out of the hood gets disinfected by spraying with 70% ethanol.
- When you're done working, reverse these steps to clean up after yourself.

In many cases, biological reagents are safe to work with at the bench. When working with organisms or biological reagents outside of the hood, remember to take sensible precautions:

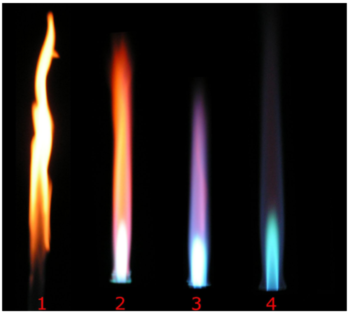


FIGURE 6: Bunsen burner flames. Source: Jan Fijałkowski. Licenced under CC-BY-SA-3.0.

- Always wear your PPE: gloves, goggles and lab coat, check!
- Use a Bunsen burner to sterilize your tools. Aim for the blue part of the flame where it's hottest (*Figure 6*).
- Wipe down the bench with 70% ethanol before and after the experiment.

Waste management

When preparing your tissue culture hood for use, make sure you've planned ahead for how to dispose of contaminated waste. Bring these items:

- a large beaker – for used tips
- a large flask with an inch or so of bleach – for discarding media
- a biohazard bin or bucket nearby – for the larger items, like used serological pipets (*Figure 7*).

Every lab will have different expectations on how to discard your biohazardous waste after an experiment. By collecting as you go, you've made everyone's job easier because the types of waste (liquid, solid, etc.) are separate and can quickly be "donated"

FIGURE 7: Biohazard waste bin. Source: Vivien Rolfe. Licenced under CC-BY-SA-2.0.

to their proper bin. Then all that's left for cleanup is the containers used to hold them. Learn about your lab's expectations with respect to biohazardous waste cleanup. It may be as simple as taking care of what you generate, though in some instances you may be responsible for moving the lab's large biohazard bin into a pickup area or autoclaving the lab's collective waste every week.

The care you take when working with cells, tissues and viruses is how you keep them growing where they belong: in a petri dish, and not anywhere else – like your stock bottle of media, in the lab, or even in you!

Radiation

You may not end up working with radiation yourself, but there's a good chance that you will work in a lab that does. While an MSDS can instruct you on how to confidently handle everyday lab reagents, radiation safety demands significantly more training and awareness. That's because mishandling a radioactive reagent can render a laboratory environment unusable and cause continuing damage should it enter your body – for years to come.

If you anticipate using radiation in your experiments, seek formal training through your organization. Before running any new experiment, review the material again, as the use of radioactivity is strictly regulated not just at the organizational level, but at the national level.

How to minimize your risk

Generally, when you're not using chemical reagents and biologicals, their hazardous potential is quite low. Radioactive items are a different beast, though, because even when they're just sitting on the bench they are constantly throwing off invisible, energetic particles.

The goal when working with radioactive materials is to use them effectively in your experiments while minimizing any potential exposure. When designing experiments that use radioactive reagents, remember the "ALARA" principle: use levels of radiation that are As Low As Reasonably Achievable.

Just like the precautions you take when you work with everyday chemicals, you can take similar steps to help reduce your risk of exposure to radiation:

- **Use protection**. Always use PPE when working with radiation, but remember that standard PPE may not be enough. Formulate a game plan before working with any "hot" items so that you understand completely what your isotope does, what it can do to you, how long it lasts (the half life) and what materials can stop it in its tracks. For example, double gloves and a thick lab coat may be sufficient protection when you use one type of isotope; for another, aluminum foil, lead or thick, dense acrylic shields may be what you need to block those particles from ever reaching your skin.

- **Work away from "hot zones"**. The farther you can work away from the lab's radiation area – and from those who are actively using it – the better the chance that there there are more solid objects between you like walls, benches and equipment to absorb any particles if they should ever escape into the lab environment. Though it is not always practical, formal exposure monitoring (see *Measure your dose: exposure monitoring*) is a good way to help assess any risk.

FIGURE 8: Radioactivity symbol. Source: **Cary Bass**. Licenced under public domain.

- **Perform fewer radiation experiments**. Fewer radiation experiments means fewer chances for exposure and less radioactive waste hanging around and decaying in the lab. It's as simple as that.

- **Design experiments intelligently**. Are you able to refine your experiment to achieve the same result with a different kind of assay that doesn't use tritium, radioactive phosphorus, carbon, sulfur or some other radioactive material? At the very least, have you explored using radioactive molecules that emit alpha particles instead of beta particles (i.e. those that decay at a faster rate)?

- **Communicate, communicate, communicate.** How do you know which tools and equipment are used with radioactive reagents? Look for yellow tape, the signal word "Radioactive" or "Radiation" and the infamous boat-screw shaped trefoil (*Figure 8*). When you are about to begin working with radioactive reagents, it's courtesy to give a heads-up to others working in the same room. That way everyone is aware and those who aren't working with radiation can plan their day – and their location – accordingly.

Measure your dose: exposure monitoring

Whether you work hands-on with radioactive materials, or just around them, you might wonder if you have ever been exposed to something, and if so, how much? Exposure monitoring programs can help you know for sure.

Begin a dialogue with your lab manager or supervisor about the specific hazards present in your lab, and consider requesting support from your organization's safety team. For radiation safety, you'll be issued a dosimeter to wear in and around experimental areas that will react with any particles if they should ever reach you. A dosimeter can provide you with peace of mind (and perhaps a sigh of relief?) and reassurance that the hazards are confined to the working area itself.

If there ever is an instance where a monitor registers exposure, you, your lab manager and the safety team will sit down to work through current procedures that guide experimentation and implement changes as needed.

Lab equipment

Lab equipment hazards stem from moving parts, electricity, doors, hinges, etc. Reading the user manual before using equipment tells you what to look out for during normal use. In this section, we'll take a look at a few types of equipment that are used regularly in the lab that can pose some (preventable) hazards.

Working with Bunsen burners

Bunsen burners (*Figure 9*) provide a hot, controlled fire for tasks like flaming the bubbles out of freshly poured agar plates or sterilizing the tops of open glass bottles. In an organized laboratory environment you don't have to worry about too much stuff being in the way, as there should be empty space above and around the Bunsen burner. In rooms with a bit more clutter, you will need to carve out an area where you can use an open flame without the fear of accidentally setting something on fire. Double-check that there is sufficient empty space around the burner – including above it! – before lighting a flame. You can also work safely with a Bunsen burner in a fume hood if there is a gas line piped in.

FIGURE 9: Bunsen burner
Source: **NASA**. Licenced under public domain.

To light your burner, turn the dial on the bottom of the burner all the way closed, then open it ½ to 2 turns to let the gas flow through it. Wait a brief moment for the gas to flow through the burner, then light it with a flint-spark lighter, being sure to keep your hand and face away from the flame when the gas combusts. The flame should be adjusted to a perfect blue. If your flame is yellow or too tall (*Figure 6*), adjust the air intake valve by turning the little metal collar at the base of the Bunsen burner tube counter-clockwise.

At the end of the experiment, turn the gas valve all the way off by turning it clockwise (closed), or so that the valve is perpendicular to the tubing, making an "L" shape. If you want to take a break during an experiment, turn off the burner. It's easy to relight it when you come back to the bench. An unattended flame is dangerous – people passing through the laboratory may not be aware that it is even lit. At the end of the day, always double-check that the valve is off before leaving the lab!

Microwaves and hotplates

Microwaves and hotplates are commonly used to heat things up in the lab and can create hazardous situations if used improperly.

If you've used a microwave at home, then you already know that too much power and too much time can make food explode. The same thing can happen in the lab with agar and gels, so pay close attention to what you're heating up before it boils over. For most lab applications, you will want to heat your item slowly on a lower power setting in short increments. Between each burst of microwaving, you can shake the bottle or tray to mix the contents and help them melt more evenly.

Hotplates are good for large volumes and heating while stirring, but they are a burn hazard. The best kind of hotplate to have in the lab is one that indicates that the surface is hot to the touch after it is turned off (though this only works while the equipment remains plugged in). If your hotplate doesn't have this function, make a practice of sensing heat with the back of your hand held an inch or two above the plate to know whether or not you should let it cool off longer.

Extreme temperatures: autoclaves, freezers and liquid nitrogen

In addition to normal health hazards like hotplates, there is also the potential to be exposed to extreme temperatures in the lab environment. The trick to working around equipment with environments that range from extreme cold (–80°C) to extreme heat (121°C) lies in wearing the correct PPE!

For the autoclave, that means donning your bright orange autoclave gloves. These are like an extra-thick lab coat for your hand because they're made of cotton, too, affording thermal protection well beyond the 121°C heat of the oven. For the low temperature freezers, thick blue gloves keep your hands dry from incidental contact with liquid nitrogen, melting ice and dry ice. Both sets of gloves fit midway up the forearm and offer good dexterity when working around these extreme environments.

Liquid nitrogen, or LN_2, is perhaps the coldest item you'll ever use in the lab. Colder than wet ice (0°C), dry ice (–80°C) or the deep freezer (–80°C), LN_2 (–196°C) is most commonly used to freeze cell and tissue samples extremely quickly. There is easily a 200+ degree difference between the liquid in the storage container and room temperature! When LN_2 comes into contact with room temperature air, it boils instantly. So how can you be sure to protect against the burning effects of cold liquid nitrogen? Keep your skin well away from contact with liquid nitrogen (even gloved hands). Wearing insulated blue gloves is a must for protecting you against incidental splashes and enabling you to use metal tools that have been cooled by contact with liquid nitrogen. Remember to wear full cover eye protection, too, in case of splashes.

When sample tubes are submerged in the liquid, the liquid may seep into them over time. Bring a tube like that back into the laboratory and the LN_2 boils, expands, and . . . surprise! The lab (and you too, if you decided not to wear PPE) is now decorated with plastic parts and potentially hazardous cells!

Exploding sample tubes isn't a common event, but the risk is very real. To reduce the risk:

- **Store vials above the liquid level**. It's still cold enough to put your samples to sleep.
- **Use tubes specifically for cryopreservation**. I used an item with a plastic gasket (or o-ring) with a top that screwed inside the tube, not over it. Please check with your lab manager to help you select the best product for your application.
- **Full-face protection is necessary**. Remember, it's easier to clean off the surface of a face shield than trying to figure out how to decontaminate your face and eyes.

When sterilizing or preserving materials at extreme temperatures, make sure to use containers that can handle the "weather". Not all plastic is created equal, and incompatible plastics can become brittle in the freezer over time. For the autoclave, using the right plastic bottle or bucket is crucial, as plastics with low temperature resistance will melt, fusing with the metal rack and sealing the drain port.

No matter what temperature you use in the lab – hot, cold, fire and ice – keep an eye on it so that it is always under your control. Use PPE for additional protection where needed, and weather the elements in an environment that is conducive for your equipment.

Chapter 4
How to handle common lab accidents

Image source: That Bill Fellow, licenced under CC BY-SA 2.0.

Even if you take every precaution, accidents do happen. In this chapter, we'll talk about how to deal with common lab accidents, and how to identify situations that are too extensive or dangerous to deal with by yourself.

The eyewash

If you have accidentally gotten any powder or liquid in your eye(s), you will need to use an eyewash immediately to flush it out. The eyewash (*Figure 10*) is a plumbing fixture near the sink – usually on a lever or hinge so that it can be swung out of the way when it's not in use. When activated, the eyewash will spray two jets of water in an arcing pattern, so all you need to do is lean your face over the sink and keep your eyes open!

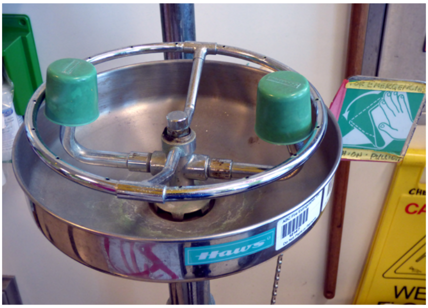


FIGURE 10: Eyewash. Source: Jason Erk.

Hopefully you'll never need to use an eyewash, but it's essential that you know where it is and how to use it, should the situation arise. Most eyewashes are paddle- or lever-activated, so give it a go and get a feel for how it works.

If the pressure is too high (it hits the ceiling when turned on!), too low (it just dribbles out) or intermittent (indicating air bubbles in the line), call in maintenance to have it adjusted. Make sure the area around the eyewash stays clear so you can swing it over the sink quickly, if necessary. Taping the area off with brightly colored tape is a great visual reminder to keep the area clear.

The safety shower

FIGURE 11: Safety shower Source: Jason Erk.

Safety showers (*Figure 11*) are used in emergencies when you have spilled a large amount of a hazardous substance on your body and/or clothing. While all labs have easy access to an eyewash, not all labs have safety shower access in the same room, as it's dependent where the plumbing is in the building.

You should know exactly where the nearest safety shower is, the way you know where the nearest exit is on an airplane. Safety showers are easy to use, as they're designed to work quickly and easily during dangerous situations – just pull down on the large handle.

Spills

At some point during your career you'll need to clean up a chemical spill. Every spill demands immediate attention because of the potential hazards, which can include anything from slips and falls at the very least to fire, flood or reactivity at the most severe. But fear not! You already have a rich knowledge of how to use most of the reagents present in the lab. And as always, the

MSDS (see *MSDSs*) is your go-to guide when mounting a safe and strategic response.

The appropriate response to a spill depends on whether it can be classified as major or minor. Examples of common minor lab spills include drips when pouring agar plates, splattered media in the tissue culture hood and scattered powder when you miss the weigh boat. Spilling a phenol-based solution would count as a major incident, as it produces a big, noxious smell that saturates the lab.

Common, minor spills:

- are generally small volumes;
- include reagents you have confidently used before;
- present no or low exposure hazard;
- require only standard lab PPE for normal handling (gloves, goggles and lab coat); and
- can be easily cleaned up on your own.

By contrast, major spills:

- are generally large volumes;
- include biologicals and radiological substances;
- present an immediate threat to your health and wellbeing;
- require more PPE than what you would normally wear at the bench; and

- require a team response to make an area safe again.

The severity of a spill incident is subjective, and can depend on how confident you are in assessing and handling the situation. When in doubt, ask for help!

Here's how to clean up most spills in 4 easy steps:

1. **Take charge**. The first step to safely handle a spill is by creating a buffer of empty space between the spill and your colleagues. For something that is relatively benign, like flooding from the distilled water generator, your colleagues can continue to work in the lab while you clean up. However, in the event of a major spill, you should encourage everyone to exit the room. That way any potential exposure is kept as low as possible. Once everyone is at a safe distance from the hazard, pull the MSDS for every chemical involved and gather the customary PPE – gloves, goggles, extra aprons and lab coats where available, paper towels, sponges and spill kits. When backup arrives, you'll be able to bring them up to speed quickly!

2. **Communicate, communicate, communicate**! Of course, Murphy's Law dictates that a spill will happen in the most inconvenient area possible, like a hallway, connecting room or anywhere in the middle of frequent traffic and far away from the PPE you need. Announce the situation so your colleagues recognize that a hazardous situation may exist, and let them know that you will give the "all clear" when it has been resolved.

3. **Contain the spill and clean it up if you are able**. One challenge with any spill is to contain it in such a way that it doesn't become a larger problem than it already is, and that others who aren't aware learn about it before walking through

it, exposing themselves. Set up a perimeter if you need to, and encourage your colleagues to leave the lab while you address the situation.

4. **Report it**. The hard fact is that every spill can be harmful. Even when you're confident in your abilities, it's a good idea to share the incident with your lab manager. For incidents where you need extra help, call in the safety people, who are specifically trained for these situations. Many institutions require you to formally report all major spills (see *Reporting accidents*), so be sure to check your organization's regulations once the situation is in hand.

Fires

Where there's *heat* (like a hot plate, or burner)...where there's a *fuel source* (lab wipes or volatile chemicals)... and where there's air for combustion (i.e. your everyday lab environment), you have everything needed to start and sustain a fire. Take any of those three things away, though, and you're well on your way to containing and extinguishing that fire. Take stock of what firefighting equipment is available to you now, and learn how to use it in case you ever have to.

FIGURE 12: Fire blanket. Source: Jason Erk.

The following are some common tools for fighting lab fires:

- **The emergency shower & lab sink**. Cold water from the emergency shower (see *The safety shower*) or sink can be used if someone has accidentally lit their clothing on fire. If you are able to quickly take off an outer layer, like a lab coat or long sleeved shirt, place it in the lab sink and run the water over it. Otherwise, hop under the shower to soak everything.

- **Fire blankets**. Extinguishing a small fire can be as simple as smothering it so that it runs out of fresh air to thrive. If your lab has a fire blanket (*Figure 12*), it will be mounted to the wall. To use it, pull it all the way out of its holder and drape it over the fire.

- **Fire extinguishers**. For large fires, and especially fires involving oils, nonpolar organic chemicals and the like, use a fire extinguisher to eliminate the immediate hazard. To combat certain fires you'll need a fire extinguisher that is filled with pressurized carbon dioxide, a chemical powder or foam. The MSDS is your key to firefighting measures. You can find fire extinguishers mounted on the wall along the corridor to your lab, near a stairwell, an exit or even inside your lab. Building fire extinguishers are marked with signs above, so look for the fire-engine red cylinder nearby.

  To use a fire extinguisher, remove it from its cabinet and pull the metal pin out of the handle. There's a plastic loop that keeps the pin securely in place so you will need to cut it off, or twist the pin a few times to break the loop before you are able to pull the pin completely out. Then it's a matter of aiming at the base of the fire, squeezing the handle and spraying from side to side. For a tutorial on how to use a fire extinguisher, see this detailed guide.

- **Alarm boxes**. Some situations are beyond your control, particularly if they are overwhelming or hazardous. For large or out of control fires, activate the building's alarm by pulling down on the pull box to alert everyone to exit the building.
  Alarm boxes are found at the end of your corridor, near exits, stairways and elevators. As an aside, pulling the alarm is also an effective way to get everyone to safety when there's a major chemical spill (see *Spills*).

The best way to fight lab fires, of course, is to take steps to prevent them from occurring. Here are 5 quick ways to reduce your risk of fire:

- **Know your firefighting measures from the MSDS**. Every chemical has different physical properties. The flashpoint and vapor pressure are two items to note particularly; the MSDS shares important information on what items – and what not to use – to fight a fire!

- **Clear space around you**. Clear the bench and shelves around your workspace to ensure that nothing accidentally hangs above or passes through an open flame while you are working (see *Working with Bunsen burners*).

- **Reagents: use working volumes**. When working with flammable reagents, rather than bring an entire 4 L carboy to the bench, pour just what you need to into a smaller secondary container. This way you're controlling how much fuel is available to a fire, should one ever start.

- **Store flammable items in the flammables cabinets**. Use the right reagent at the right time. When flammable reagents are stored in the cabinet, you've physically removed them from active work areas and placed them behind a protective shield.

- **Check the lab before you leave at the end of the day.** Before you leave the lab, check that the gas valves are closed, the hot plate is cool, off and unplugged and flammable reagents are stored in the flammables cabinet. This is a proactive step that will help make the lab safe overnight.

Reporting accidents

For all the safety measures implemented in research labs, accidents still happen. If an accident does occur, how do you report it so that steps can be taken to ensure that the same event never happens again?

Each lab and/or organization has defined criteria for what accidents need to be formally reported. Look for this information during your safety training, in your employee handbook or a nearby emergency response flipbook. It's best to report any accident immediately if you are able, otherwise as soon as reasonably possible.

In general, you must always report situations that:

- Result in fire, flood or blood.
- Result in illness.
- Involve a physical injury, burns, or exposure (however minor).
- Involve chemicals with a high health, flammability or reactivity hazard.

- Involve compressed gas cylinders.
- Involve sharps.
- Involve biohazardous materials or radiation.
- Cause any type of spill.

If you spot a potentially dangerous situation that could create a “perfect storm”, report it informally to your lab manager. This includes obvious situations like frayed wiring, equipment that trips the circuit breaker and nearly-full waste containers. Other potentially dangerous situations that are not so obvious include leaky carboys stored on a shelf above eye level or open buckets of mystery liquid sitting on the floor of the walk-in cooler.

Reviewing accident and injury reports will help you avoid similar accidents in the future!

Chapter 5
Conclusion: Great places to find more safety information

Image source: Magnus Akselvoll, licenced under CC BY 2.0.

As we mentioned at the beginning of this book, this is a simple BitesizeBio introduction to lab safety. There are certainly many topics that we haven't covered, including an equal number of interesting topics that you may now want to learn more about. Fortunately, there are a ton of great resources that are available to point you in the right direction!

Your lab's safety manual

Each lab has an action plan entitled the "chemical hygiene plan" or "laboratory safety manual." This is your how-to guide for best work practices. Most questions about lab safety at your organization can be answered by this document. Look for it next to your lab's collection of MSDSs.

Where the chemical hygiene plan ends, a laboratory may compose appendices to cover specific items used in their research. Alternatively, this may be a separate document penned by your lab manager (i.e. the "lab rules").

Your institution's safety department

There are also people outside of your lab who care about your environmental safety and health, and are always on call. Turn to the safety department to ask a question when you need an answer right away. These people know policy and regulation like the back of their hand, and can share information in a way that makes sense. The safety department is also in charge of overseeing annual training, which is a great source of info for general lab safety, biological safety and radiation safety. At most institutions, you'll do this training when you join a lab and then every year after that.

Your right to know: the Hazard Communication Standard (HCS)

Another good document to reference is the Hazard Communication Standard (HCS). The Hazard Communication Standard empowers you by providing valuable information on how to recognize which chemicals in your inventory are toxic or hazardous, and helps you understand any precautionary measures that you need to take before using them. It's the rulebook that chemical manufacturers and employers play by, so that you have the right tools for the right parts of your job. Communication and training are big parts of the HCS. Very simply, the HCS guarantees you the right to know about hazards that can affect you in the lab. You can read more about the Hazard Communication Standard here.

The Globally Harmonized System of Classification and Labeling of Chemicals (GHS)

The Globally Harmonized System (GHS) for labeling chemicals makes hazard identification easy: commonly recognized pictures, words and symbols call your attention to the inherent hazards with your reagents, allowing you to assess and protect against them. You can read more about the GHS here.

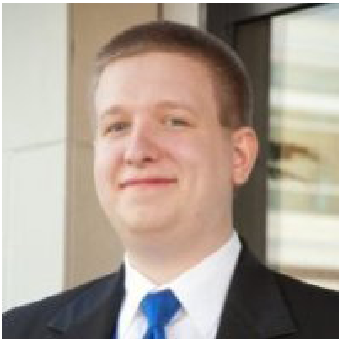
For even more information, check out these other great resources:

- Enforcing Hazard Communication Standards
- HazCom FAQ
- GHS (Globally Harmonized System) Answer Center

- US Department of Health and Human Services: Enviro-Health Links - Laboratory Safety
- OSHA Laboratory Safety Guide
- Hazard Communication Standards Pictograms

At the end of the day, it all boils down to this: the more knowledge you have at your fingertips, the better prepared you are to deal with potential lab hazards. And that enables you to reduce hazards in some areas and eliminate them in others, empowering you to conduct research in the safest manner possible!

About the author:
Jason Erk



Jason holds a BSc in biochemistry and is a faculty research assistant in a university laboratory, where related projects span the fields of behavior, genetics, neuroscience and addiction research. Since 2011, he's contributed to BitesizeBio on a variety of interesting and diverse "how-to" life science topics. Follow along with him here , or connect on LinkedIn.