

Basics of laboratory safety

Learning Outcomes

- What is laboratory safety?
- Chemical and Biological Safety.
- Control measures to be adopted.
- Good Laboratory Practices- Do's and Don't's
- Classification of Biosafety laboratories and their specific requirements.
- Signage

Hazardous chemicals

Routes of exposure

Inhalation

Most serious

Most readily
absorbed

Gases, fumes,
vapours, very
fine powders

Skin contact

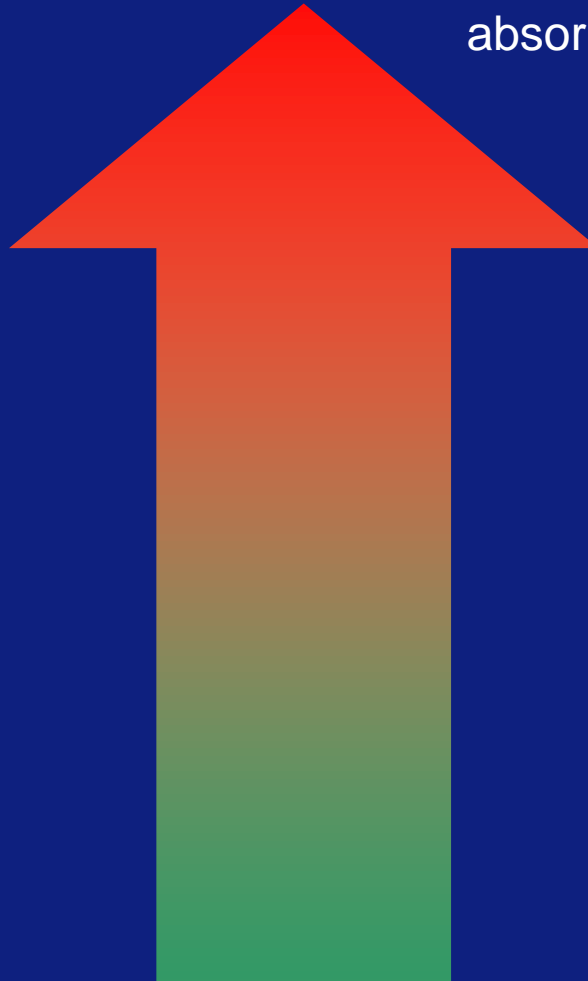
Corrosive
liquids and
those that
readily cross
the skin barrier

Ingestion

Least likely

Least readily
absorbed

Dense solids



Risk factors

Intrinsic hazard associated with the substance

Severity of hazard



Low health hazard

Substances designated as irritant (those that affect the skin and sometimes the eyes)



Medium health hazard

Substances designated as corrosive, irritant (particularly by the inhalation route) or harmful (particularly by the inhalation route)



High health hazard

Substances designated as toxic, very toxic, carcinogenic, mutagenic and those that may impair fertility or harm the unborn child

Risk factors

Risk Phrases

- All hazardous substances are allocated Risk Phrases that describe the particular hazard(s) associated with that substance
- Risk Phrases can be found on the Material Safety Data Sheet (MSDS) supplied with the substance.
- It is a legal requirement for suppliers to provide safety information in the form of an MSDS

15 - Regulatory Information

CLASSIFICATION AND LABELING ACCORDING TO EU DIRECTIVES

ANNEX I INDEX NUMBER: 602-006-00-4

INDICATION OF DANGER: Xn

Harmful.

R-PHRASES: 22-38-40-48/20/22

Harmful if swallowed. Irritating to skin. Limited evidence of a carcinogenic effect. Harmful: danger of serious damage to health by prolonged exposure through inhalation and if swallowed.

S-PHRASES: 36/37

Wear suitable protective clothing and gloves.

Risk factors

Risk phrases

R1	Explosive when dry
R2	Risk of explosion by shock, friction, fire or other sources of ignition
R3	Extreme risk of explosion by shock, friction, fire or other sources of ignition
R4	Forms very sensitive explosive metallic compounds
R5	Heating may cause an explosion
R6	Explosive with or without contact with air
R7	May cause fire
R8	Contact with combustible material may cause fire
R9	Explosive when mixed with combustible material
R10	Flammable
R11	Highly flammable
R12	Extremely flammable
R14	Reacts violently with water
R14/15	Reacts violently with water, liberating extremely flammable gases
R15	Contact with water liberates extremely flammable gases
R15/29	Contact with water liberates toxic, extremely flammable gases
R16	Explosive when mixed with oxidising substances
R17	Spontaneously flammable in air
R18	In use, may form flammable/explosive vapour-air mixture
R19	May form explosive peroxides
R20	Harmful by inhalation
R20/21	Harmful by inhalation and in contact with skin
R20/21/22	Harmful by inhalation, in contact with skin and if swallowed
R20/22	Harmful by inhalation and if swallowed
R21	Harmful in contact with skin
R21/22	Harmful in contact with skin and if swallowed
R22	Harmful if swallowed
R23	Toxic by inhalation
R23/24	Toxic by inhalation and in contact with skin
R23/24/25	Toxic by inhalation, in contact with skin and if swallowed

Currently 60+ Risk
Phrases listed under
Chemical (Hazard
Information and
Packaging for Supply)
Regulations
(CHIP)

Risk factors

It is also a legal requirement for suppliers to provide appropriate hazard labelling for chemicals

How many types of hazard label are there?



Packing and Supply



Hazard Warning







Transport




Risk factors

Suppliers must also identify and label chemicals that present a safety risk as opposed to a health risk.....

Symbol	Abbreviation	Hazard	Description of hazard
(Physicochemical)			
	E	explosive	Chemicals that explode.
	O	oxidising	Chemicals that react exothermically with other chemicals.
	F+	extremely flammable	Chemicals that have an extremely low flash point and boiling point, and gases that catch fire in contact with air.
	F	highly flammable	Chemicals that may catch fire in contact with air, only need brief contact with an ignition source, have a very low flash point or evolve highly flammable gases in contact with water.

....and those that have an environmental impact

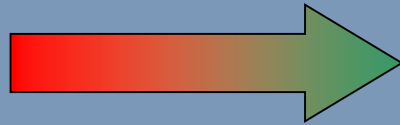
(Environmental)			
	N	dangerous for the environment	Chemicals that may present an immediate or delayed danger to one or more components of the environment

Control measures

Prevention of exposure takes priority over control

Substitute the hazardous chemical with a safer alternative

**Ethidium
Bromide**



Use a safer form of the same chemical

Exclude non-essential personnel from the area

- Access control systems
- No write up areas in labs
- Hazard zoning



Control measures

Minimise the quantities used, stored and transported

Ventilation:
use fume cupboards
where necessary.....
and check that they
are being maintained



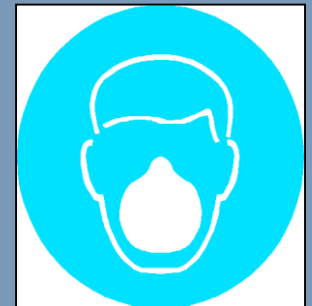
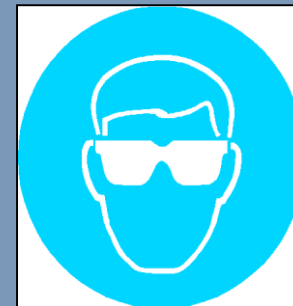
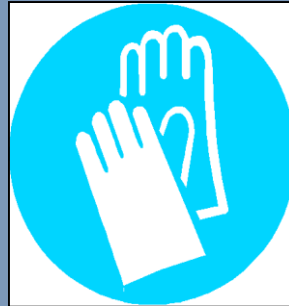
Store hazardous chemicals
in an appropriate manner

Control measures



Transport hazardous chemicals using appropriate containment methods

Wear appropriate Personal Protective Equipment (PPE)



Dispose off chemical waste properly: sink or specialist waste contractor?

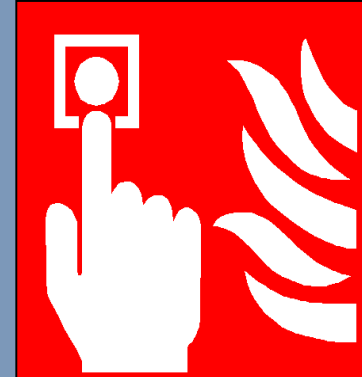
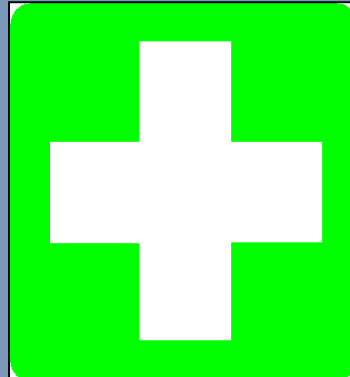
Control measures

Observe
good
housekeeping



Ensure users are provided with sufficient information,
instruction, training and supervision

Ensure that suitable emergency procedures are in place



No horseplay in the lab !!!!!!!!!!!!!!!!!!!!!



Laboratory activities that present a specific risk

Activity	Control measures
Weighing	Consider using fume cupboard or other benchtop enclosure such as weighing station
Pipetting	Consider using fume cupboard and or drip trays; dispose of pipette tips properly; use good pipetting technique; clean and maintain automatic pipettes
Shaking / mixing	Consider using fume cupboard in conjunction with PPE
Centrifugation	Ensure compatibility of tubes with chemicals; balance tubes; PPE; clean-up procedures and training
Syringe filtering	Select right pore size; dilute sample; PPE
Use of sharps	Eliminate wherever possible; select right tool for job; wear appropriate PPE; ensure safe disposal
Elevated temperature	Lower temperature if possible: use fume cupboard; cover or partially cover; reduce surface area
High pressure equipment	Consider location of equipment in conjunction with PPE

Accidents do happen.....

- Chemical splash to face / eyes
- Chemical splash to hand / arm
- Chemical splash on laboratory coat
- Chemical injected beneath skin by needle
- Exposure by inhalation (sometimes including eye irritation)
- Numerous reports of 'solvent' smells
- Allergic reaction attributed to chemical contact
- Acute asthma attack
- Spillage onto bench / floor
- Chemical in mouth – rare

....and a skin blister arising from self treatment of wart on foot with liquid nitrogen!

Sources of further information

DAFF (Department of Agriculture, Forestry & Fisheries)

<http://www.daff.gov.za/daffweb3/Search-Results-Page?Search=biosafety>

Sigma Aldrich

http://www.sigmaaldrich.com/Area_of_Interest/Europe_Home/UK.html

Wikipedia:

<http://en.wikipedia.org/>

SCENARIO:

You enter a laboratory first thing in the morning and notice an odour. You recognise that two specimen jars on the benchtop have cracked and spilled 4% formaldehyde onto the benchtop and an area of the floor. There is still evidence of liquid present.

WHAT DO YOU DO NEXT?

MODEL ANSWER

1. Leave the room again immediately.
2. Close the door and secure if possible.
3. If the door cannot be secured, attract the attention of a colleague to stand by the door or prepare some temporary warning signage to affix to the door.
4. Report the incident to your Group Safety Adviser / Safety Officer.
5. Nobody should enter the room until the airborne concentration of formaldehyde is established. This can be determined using a portable formaldemeter – several meters are kept by the Safety Department and others are held locally.
6. Once the readings have been established there are two options:
 - a. A member of staff can enter wearing suitable respiratory protective equipment and clean the spillage up using a polymerising agent.
 - b. The room can be left vacant until the formaldehyde has evaporated completely. The length of time that this takes will be dependent upon the quantity spilt and the ventilation standards within the laboratory.
7. Complete a dangerous occurrence report in accordance with established University procedures.

What is Biosafety?

- Safety from exposure to Infectious Agents

Smallpox



Biosafety in Academic Research

- Research Universities:

Promoting safe laboratory practices, and procedures; proper use of containment equipment and facilities; provides advice on laboratory design and risk assesment of experiments involving infectious agents, rDNA in-vitro and in-vivo.



Biohazard Symbol

Bottom Line: Risk & Containment



Biosafety Issues

- Laboratory Safety
- Bloodborne pathogens (BBP)
- Recombinant DNA (rDNA)
- Biological waste disposal
- Infectious substance and diagnostic specimen shipping



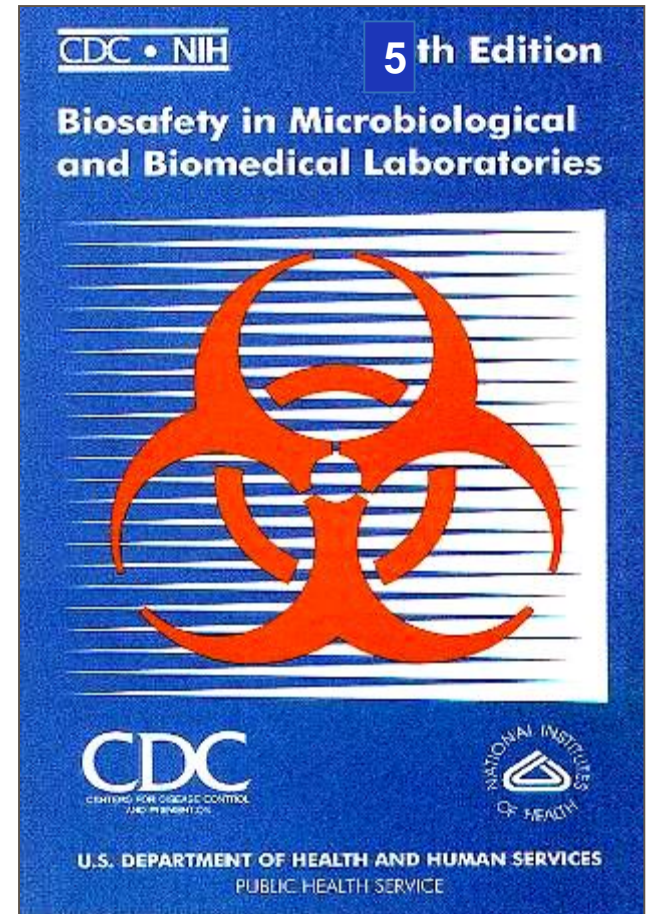
Biohazardous Materials

- Viruses
- Bacteria
- Fungi
- Prions
- Recombinant DNA
- Transgenic Plants, Animals and Insects
- Human and Primate Cells, Tissues, and Body Fluids

Biosafety Concepts

Biosafety In Microbiological and Biomedical Laboratories

- “BMBL” (acronym)
- CDC/NIH Publication
- Safety “Guidelines”
- Regulations of Institution receives NIH funding
 - Code of Practice and “Gold” Standard in Industry
- Clinical & Research Lab.
- Lab. Animal Facilities



Biosafety Concepts

Principles of Biosafety

- Practice and Procedures
 - Standard Practices
 - Special Practices & Considerations
- Safety Equipment
- Facility Design and Construction
- Increasing levels of protection





Principles of Biosafety

Biosafety Levels 1-4 (BSL)

- Increasing levels of employee and environmental protection
- Guidelines for working safely in research & medical laboratory facilities

<https://consteril.com/biosafety-levels-difference/>

Biosafety Level-1

Concepts of Biosafety

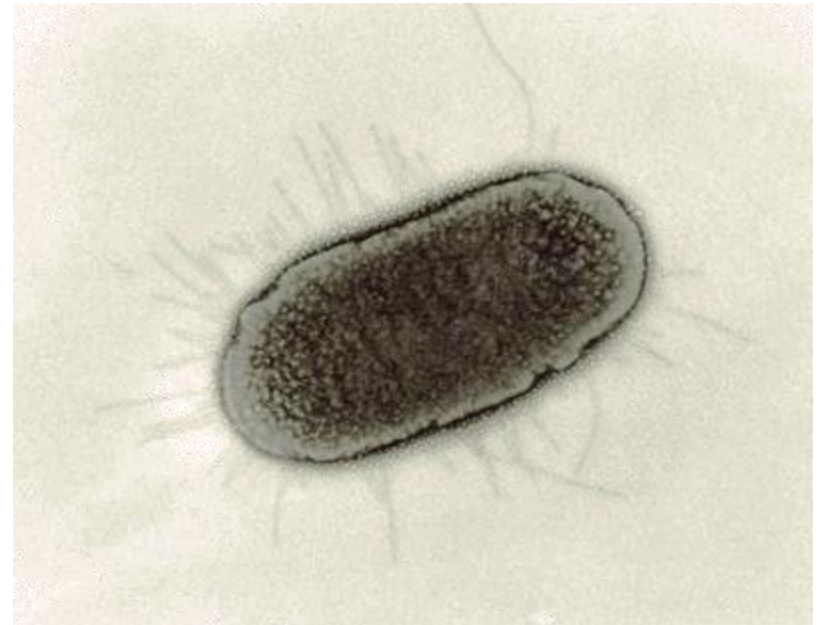
Biosafety Level-1 (BSL-1)

- Well characterized agents
- Agents not known to cause disease (in healthy human adults; now healthy immunocompetent adults)
- Prophylactic treatment available
- Open bench procedures
- Animals in open cage system or open environment (outdoors)
- Good laboratory practices



Risk Group 1 Agents

- E.coli K-12
- Transgenic Plants
- Plasmids
- Fungi
- Mold
- Yeast



BSL-1 Practices

- Bench-top work allowed
- Daily Decontamination
- Required Handwashing
- Biosafety cabinet not required
(unless creating aerosols)
- Do not require biosafety symbol display





Risk Group 2 Agents

- Human or Primate Cells
- Herpes Simplex Virus
- Patient specimens
- HIV



BSL-2 Practices

Concepts of Biosafety

Practices & Procedures

- Agents associated w/ human disease
- Treatment for disease available
- Agent poses moderate hazard to personnel and environment
- Direct contact or exposure
- Percutaneous exposure
 - Scratch, Puncture, Needle
 - Mucus membrane exposure
 - Eyes, Mouth, open cut

BSL-2 Practices

- Limited access to lab when work in progress
- Daily decontamination
- Labcoat, safety glasses and gloves required
- Red bag & sharps containers required
- **Require display of Biosafety symbol**



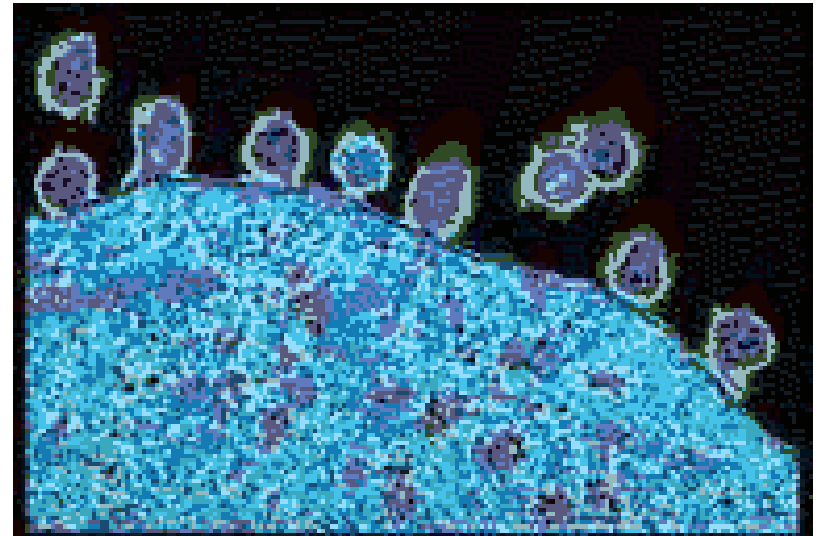
BSL-2 Practices (con't)

- Label all equipment (incubators, freezers, etc.)
- TC room – negative air flow
- Documented training



Risk Group 3 Agents

- *Mycobacterium tuberculosis*



Biosafety Level 3

Working in High Containment

Biosafety Level-3 (BSL-3)

- Indigenous or exotic agents
- Aerosol transmission
- Serious health effects
- Treatment may or may not exist



BSL-3 Practices

- Public access NOT permitted
- Daily decontamination after spill and upon completion of experiment
- Autoclave required and waste is disposed at the end of day
- Required foot activated handwashing sink and controls
- No sharps unless absolutely necessary



BSL-3 Practices (con't)

- Aerosol minimization procedures required
- Wrap around disposable clothing is required. Specialized equipment may be required depending upon procedures
- **Biohazard Signs and labels posted**





BSL-3 Practices (con't)

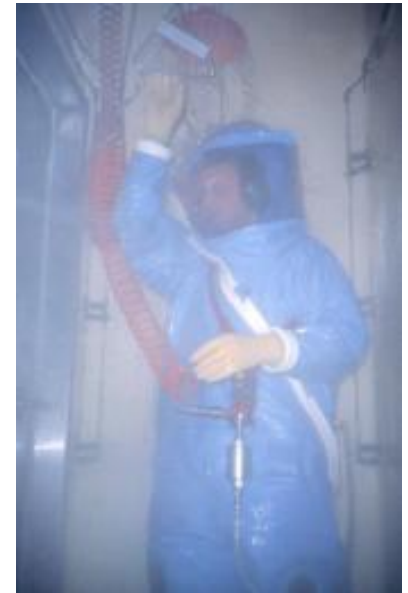
- Bench top work not permitted
- Documented training and personnel competency certification (for BSL-3 procedures)
- Spills – report immediately and treat accordingly
- Vaccinations/post exposure protocols and SOP's, Biosafety Manual, Biosafety Officer

Biosafety Level-4

Working in High Containment

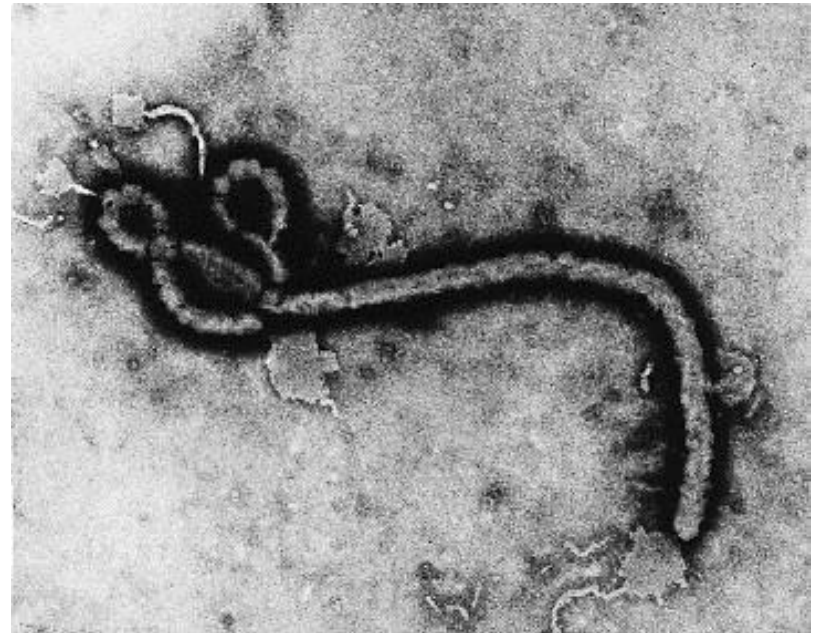
Biosafety Level-4

- Builds on BSL-3 practices
- Maximum containment facilities
- Pressurized Containment Suite
 - BSL-3 + Class III Biosafety Cabinet
- Chemical decontamination showers
- Liquid effluent collection / decontamination



Biosafety Level 4

- Lassa Fever Virus
- Ebola Hemorrhagic Fever Virus
- Herpes B Virus



Biosafety Concepts

Working in High Containment

Biosafety Level-4 (BSL-4 or ABSL-4)

- Dangerous/exotic agents
- Life threatening disease
- Aerosol transmission
- Agents of unknown risk of transmission or health affects
- No known treatment





Addressing Risk Assessments

- What is the organism?
- Is it Wild-type or chemically treated?
- What is the max. concentration, volume, infectious dose?
- What is the work space like?
- Aerosolizing procedures? How do they contain their aerosols?

Risk Assessment, con't

- Are personnel trained? Do personnel understand the organism, infectious dose and symptoms?
- What are their experimental procedures?
- Will they be transporting the material? Shipping intra, inter-state or international?
- Are they doing tissue culture?
- Do they have adequate containment equipment?



Risk Assessment, Con't

- Are they doing this work in-vivo? Have you consulted and discussed this with the Vets and Animal unit to determine special needs and housing?
- Waste issues addressed?
- Pregnancy issues with the organisms?



First Aid Measures



- Splash to Eye
 - Rinse thoroughly for 15 minutes at the eyewash or sink



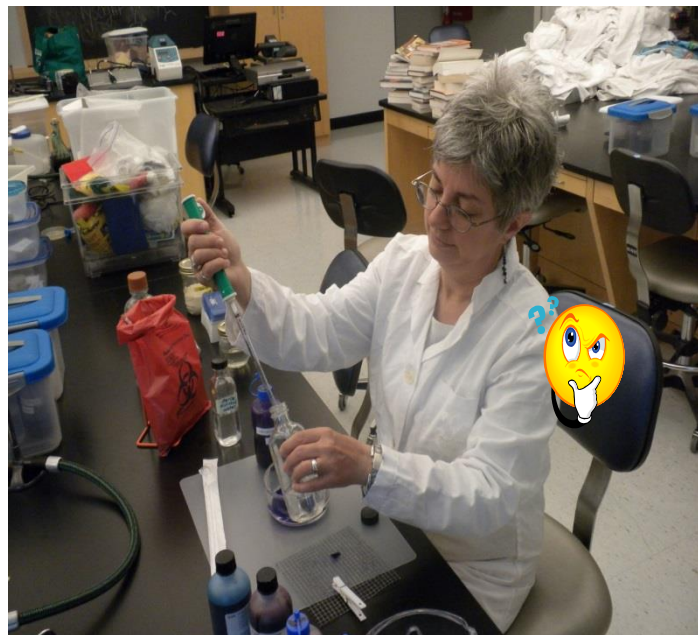
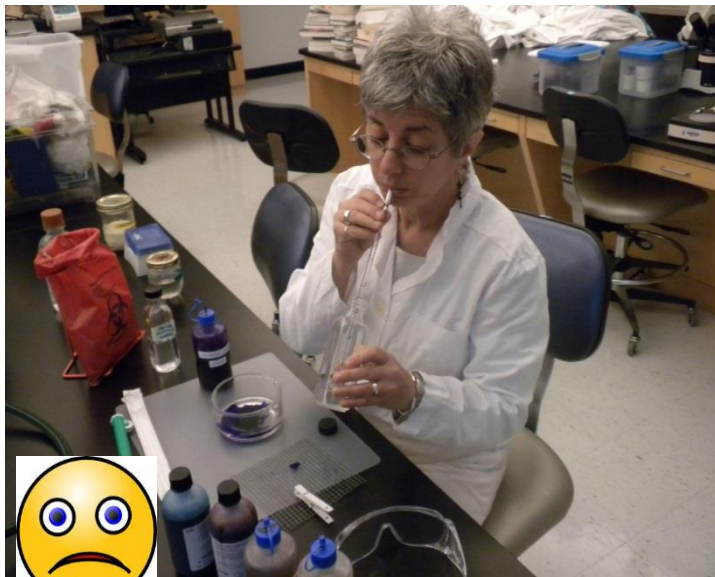
GOOD LABORATORY PRACTICES





Gloves?

Lab coat... must be worn properly



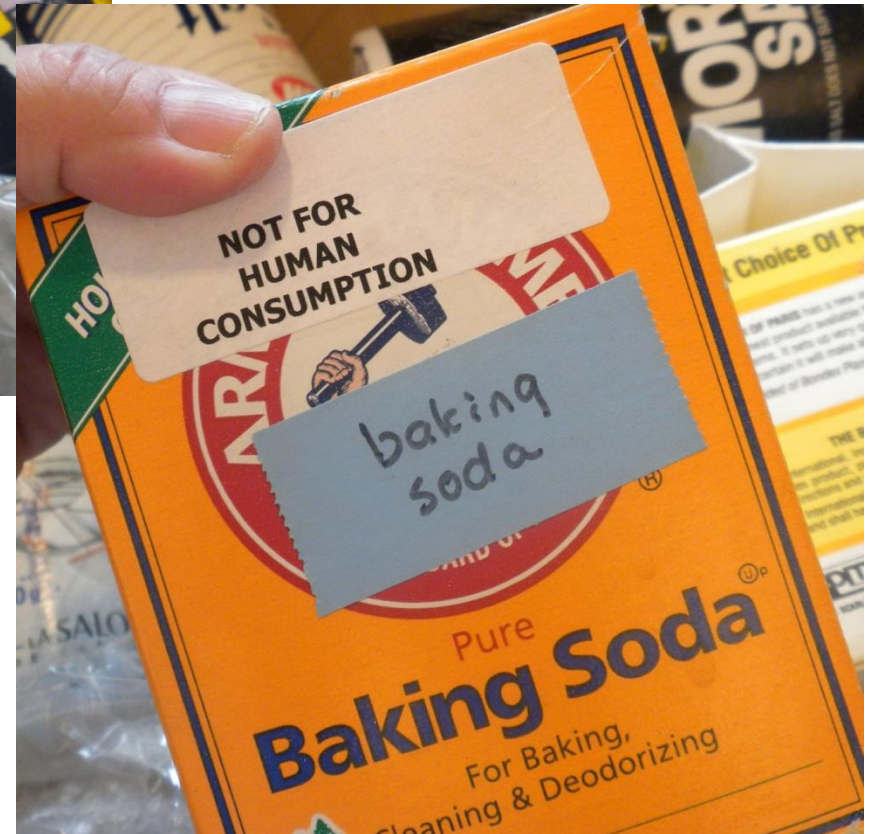
Mouth pipetting? Goggles.



Labeling of chemicals and solutions is important.



Anything normally found in a kitchen, or thought of as “food” needs to be labeled otherwise.





Biological waste must be disposed of properly.



HAND HYGIENE



Resources

- UCSD Biosafety: <http://blink.ucsd.edu/safety/research-lab/biosafety/>
- Biological Safety MSDS: <http://www.phac-aspc.gc.ca/msds-ftss/index-eng.php>
- NIH BMBL:
<http://www.cdc.gov/od/ohs/biosfty/bmbl5/bmbl5toc.htm>
- Wits
<https://www.wits.ac.za/research/about-our-research/ethics-and-research-integrity/institutional-biosafety-committee-ibc/>

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<http://wits-enterprise.co.za/>