

IISER TIRUPATI SAFETY MANUAL



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Importance of Safety

Safety and health considerations are as important as any other materials taught in science curricula. Occupational injury data from industry studies indicate that the injury rate is highest during the initial period of employment and decreases with experience. Therefore, it is essential that everyone working in any kind of organization is aware of what can go wrong while working, how to prevent such events from occurring and what to do in case of an emergency.

IISER Tirupati is committed to ensuring the health and the safety of every individual working in the institute. We strive to make the necessary arrangements for everyone's safety within the institute. However, no safety measures are of use unless there is active cooperation of the individual under concern. Therefore, we urge everyone to follow the safety policies and regulations of the institute given below.

Fire Safety

Fire is the most common safety hazard in any organization. Therefore, it is very essential for everyone to know how to survive a building fire and what to do in case of a fire. The following section briefly explains some common protocols and procedures that may be followed during a fire emergency.

How to Survive a Building Fire

- ➤ Go out of the building immediately through the nearest exit.
- Always use stairs, not an elevator
- > Close doors from outside in case of severe fire to prevent the fire from spreading
- > In case of heavy smoke, crawl low and if possible tie a wet cloth on your nose
- > Use a fire extinguisher, if the fire is very small and you know how to use it safely

If you are on fire - Stop, Drop and Roll

If you get trapped:

- Close the door
- > Open the windows if safe
- > Do not jump out of a tall building
- Signal for help and call 101



Fire Extinguishers:

Fire, depending on its origin, is classified as:

- 1. Class A: Combustible materials: Wood, paper, furniture etc.
- 2. Class B: Flammable Liquids
- 3. Class C: Flammable Gas
- 4. Class D: Metal Fire
- 5. Class E: Electrical equipment
- 6. Class K: Oil, greases

If FIRE occurs:

It may not be necessary to evacuate the building for a small fire. If, however, there is any chance that the fire may endanger others or may cause serious damage, confine or control the fire only if possible.

Use an appropriate extinguisher:

- Dry Powder (for all type of fire): Dry powder containing extinguishers are recommended for putting off all types of fire.
- CO₂ (for B and C type fire): Because the use of dry powder leaves a messy surrounding, CO₂ based extinguishers are often recommended for small fire of chemical origin.
- If fire breaks out close to a flammable gas supply or close to electrical power source, turn off gas supplies and electrical power sources.

If a solvent in a beaker catches fire, covering the beaker and depriving the fire of oxygen can easily extinguish the fire than using a fire extinguisher on the same beaker, which may cause the solvent to spill, thus increasing the hazard!

Immediately after a fire extinguisher has been used, make a report of it and inform the Safety Officers.

If Emergency Occurs: Pull The Fire Alarm And Evacuate The Building.

If there are injured victims, provide the minimum necessary first aid '*Only If You Are Sure That There Is No Danger To Yourself*'. If providing assistance will endanger you, *DO NOT* attempt intervention and move the victim immediately to the nearest hospital. In case of urgency, call:



- 1. Ambulance 102
- 2. Fire station 101
- 3. Disaster management 108
- 4. A.P. State Emergency Service And Fire Station, Tirupati 0877 226 0101

The above list is not complete and there could be many other scenarios, not listed here. In such cases your response should be based on the given scenario.

Electrical Safety

Should you be concerned with high voltage or high current? In fact, it is a bit of both! If the voltage is not high enough, it may be relatively safe to touch an electrical circuit which can deliver high current. On the other hand, if the voltage is very high, but the current that the supply can deliver is very low, you might still be safe. Let us see why! The electricity is nothing but the flow of charged particles. In most of our everyday life situations, it is the flow of electrons. The electrons flow from a high potential to a lower potential. What happens when you get a shock is that the electricity flows through your body from an electrical circuit carrying a high potential to the ground.

Your body has certain electrical resistance. From the tip of your finger to your feet, it is about 100 k Ω under normal circumstances. However, if you are wet, the resistance can drop to about one kilo-ohm! Thus, if you are touching a 100 V terminal, the current that can flow through your body is about one milliamp under normal conditions. The resultant shock is barely perceptible (See the table below)! However, once the current passes through the body, its resistance decreases and more current starts flowing, which is indeed dangerous. If your body is wet, the initial current can be up to 100 mA, which is indeed fatal! However, if the power supply can deliver a maximum of one milliamp of current, and if it is not faulty, you are very likely safe! If there is some failure in the grounding of the power supply, the resultant current can be lethal! Therefore, do not touch any electrical terminals that look suspicious.

One important thing to note at this point is that once the electricity starts flowing through your body, your resistance will decrease drastically facilitating more current to flow. Given below in a box is a description of how human body responds to various amounts of currents [Source: Fish, R. M. & Geddes, L. A. Conduction of electrical current to and through the human body: a review. Eplasty 9, e44 (2009).]





1 mA	Barely perceptible
16 mA	Maximum current an average man can grasp and "let go"
20 mA	Paralysis of respiratory muscles
100 mA	Ventricular fibrillation threshold
2 Am	ps Cardiac standstill and internal organ damage
15/20 Am	ps Common fuse or breaker opens circuit*

*Contact with 20 milliamps of current can be fatal. As a frame of reference, a common household circuit breaker may be rated at 15, 20, or 30 amps.

Lethal voltages present in the labs can be identified by the following symbol.



Electricity can kill. Respect it!





Be Safe & Enjoy Science

First Edition

IISER Tirupati



Introduction:

Safety and health considerations are as important as any other materials taught in science curricula. Occupational injury data from industry studies indicate that the injury rate is highest during the initial period of employment and decreases with experience. Working with chemicals poses continuous challenge to one's health and to the environment. The requirements and recommendations of this 'Laboratory Safety manual' will not fully protect you unless you exercise diligence in your daily work, or at least stop periodically to assess your environment. Step back and look carefully at your laboratory environment, looking at it as a first-time visitor would. Does it look safe, neat, and orderly? Are chemicals stored properly? Are you and other personnel taking appropriate precautions? Can you see ways to make the lab safer? You are strongly encouraged to conduct assessment of your laboratory's safety practices at least once in six months (and preferably more frequent). Following certain discipline and working as a responsible human being, one can maintain a total hygiene working with chemicals. Below are few guidelines to ensure maximum safety of the organization (IISER fraternity and its properties) as well as of our society.

1. General Laboratory Protocols

1.1 Basic Rules

- > Safety goggles are mandatory for all the lab workers
- Lab workers should not work alone inside the laboratory
- > Lab-coats are not allowed in common areas like cafeteria, restrooms etc.
- Lab worker should be aware of the location and proper operation of laboratory safety equipment
- > Know the exits in the laboratory and in the building
- > Everyone while working in the lab should wear lab coats and full pants.
- Use of blast shields is strongly recommended while doing potential dangerous reactions (such as dealing with peroxides, diazo-compounds, high pressure vessels, distillation of high boiling substances).
- Gloves shall be worn while working in the lab and should be removed before touching surfaces outside the work area (i.e., doorknobs, computers etc).
- > Feet should be covered completely with shoes containing reasonable heel heights.
- > Open toe sandals and half pants/shorts should be avoided in the lab.

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1.2 Good Personal Habits & Behavior

- > Students should act in a professional manner at all times.
- > Eating, drinking, gum and tobacco chewing, are not permitted in the laboratory.
- ▶ Using mobile phone is strictly prohibited inside the laboratory.
- Ice from the ice machines for laboratory use shall not be used for beverages, food or food storage.
- > Do not smell or taste chemicals.
- > Skin contact with chemicals should be avoided.
- ➤ Hands should be washed thoroughly before leaving the lab.







1.3 Housekeeping

- ▶ Lab areas are to be kept clean and uncluttered.
- > Spills should be cleaned up immediately from work areas and floors.
- Equipment and instrumentation shall be cleaned to remove spillage and contamination before repair or calibration service is requested.
- Personnel must be able to see clearly through the protective glass sashes on fume hoods.
- Clear aisles, exits, and hallways for obstructions leading to slipping or tripping hazards (e.g., boxes, electrical cords or other items on the floor).
- > Ensure unblocked access to all of the following:
 - Eyewash/safety showers
 - Electrical panels
 - Fire extinguishers
 - Chemical storage cabinets
 - Fume hoods
 - Waste containers

1.3.1 Possible ways to avoid clutters in Labs

- Laboratories that are cluttered present a variety of safety hazards to researchers, students, and visitors. Some common methods for controlling clutter in laboratories are as follows:
- Properly dispose of chemicals and equipment that are no longer needed
- Do not buy chemicals, solvents or other inventories in bulk unless there is space available to safely store the material.
- Regularly schedule "lab clean-up days"
- Remove clutters from fume hoods, and ensure that they are not used for longterm storage of equipment, chemicals, or supplies that are not regularly used in the fume hood.
- Empty containers of unwanted materials (including trash) on a regular basis, and never allow them to overflow.
- Store excess materials in a neat, secure manner that provides easy access and reduces the potential for falling, collapsing, rolling, or spreading of the material.
- Limit overhead storage to lightweight, non-hazardous items.
- Chemical containers, supplies, and equipment are to be stored away from the edges of benches and shelves.
- Never stack chemical containers directly on top of one another (unless in original boxes that can be safely stacked) and/or with incompatible chemicals (such as acids with bases or flammables with oxidizers).

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- Containers holding chemicals should not be stored on the floor. When this is unavoidable, store containers in plastic tubs or other secondary containment.
- Clearly mark bench areas containing radioactive materials with radiation tape and sorbent pads.
- Clean up all spills promptly. Never leave puddles, powders, or unknown materials on floors or work surfaces.
- Daisy-chaining of extension cords and/or power strips is not permitted. Store equipment, chemicals, glassware, and supplies not in regular use away from workstations.
- Do not use leg space beneath benches and desks in a way that prevents proper ergonomic posture.
- Keep personal desk spaces and other araes clean (free of all hazardous research materials).
- Neatly store lab coats and safety glasses away from potential sources of contamination.
- Never store hazardous materials in refrigerators that contain food.
- Only eat or store food in designated areas. Maintain sufficient open space within the laboratory to manage the acquisition and disposition of materials.





1.4 GHS (Globally Harmonized System)

GHS stands for the Globally Harmonized System of Classification and Labelling of Chemicals that defines and classifies the hazards of chemical products, and communicates health and safety information. The goal is that the same set of rules for classifying hazards, and the same format and content for labels and safety data sheets (SDS) will be adopted and used all around the world. It is important that those working with chemicals are aware of all the signs and symbols on the chemical containers. The following is the symbol and related hazard information that are commonly encountered in research labs.





2. Chemical Procurement & Storage

2.1 Procurement

All material safety data sheets (MSDS) of non-catalogue compounds that are received with shipments to the lab are maintained on file. For regular catalogue items (e.g. Aldrich, Alfa Aesar, TCI etc.) the corresponding supplier home page is consulted.

2.2 Hazardous Chemical Inventory

Each laboratory should annually conduct and document hazardous chemical inventory. While no standardized format is required, the inventory should include, at minimum: the chemical name, container size, and the room number. Unused or unwanted chemicals should be disposed.

2.3 Storage

- Stored and working amounts of hazardous chemicals shall be kept to a minimum.
- Bulk store (in liters quantity) place for flammables should be away from working laboratory.



- > All containers must have a legible and firmly attached label
- > Compressed gas cylinder must be supported using straps, chains, or stands
- Acids, bases, flammables, strong reducing and oxidizing agents should be segregated within the laboratory. Water reactive materials must be separated from all other chemicals.



3. Electrical Safety

Electrical equipment and wiring comprises a major part of the laboratory, thus posing a new set of possible laboratory hazards. Periodic laboratory inspections should pay particular attention to electrical safety.





A list of possible wiring hazard are as follows:

- Spliced cables
- Worn-out cables
- Tripping hazards from poorly draped cables near hot plates etc.
- Sliced cables near sinks or other wet locations.

4. Fume Hood Safety and Ventilation

General laboratory ventilation shall provide airflow into the laboratory from nonlaboratory areas and out to the exterior of the building. Laboratory doors should remain closed, except for exit and entrance. All reactions must be performed within a fume hood. The hood sash should remain closed or at minimal safe height while working in the lab. Ventilation problems or fume hood alarms should be reported to the concerned lab supervisor who shall submit repair requests to facilities maintenance.

5. Waste disposal:

Ensure proper segregation, containment and storage of wastes. Liquid wastes are to be segregated according to halogenated, non-halogenated and aqueous wastes. Solid wastes are to be segregated as silica waste, heavy metals (palladium, Ra-nickel) waste and miscellaneous other contaminated solids wastes such as papers, cottons, gloves, facemask etc. Disposable plastic syringes and plastic chemical containers should be collected separately. Sharp objects like syringe needles and broken glass pieces (Chemically contaminated broken glassware's should be segregated) should be separately collected in appropriate containers. The waste containers should be labeled properly and disposed through an agency. Give special attention to avoid mixing of incompatible wastes to minimize disposal costs.

6. Fire Safety

Fire is the most common safety hazard in any organization. Therefore, it is very essential for everyone to know how to survive a building fire and what to do in case of a fire. The following section briefly explains some common protocols and procedures that may be followed during a fire emergency.

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- > In case of heavy smoke, crawl low and if possible tie a wet cloth on your nose
- > Use a fire extinguisher, if the fire is very small and you know how to use it safely

If you are on fire - Stop, Drop and Roll

If you get trapped:

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If FIRE occurs:

It may not be necessary to evacuate the building for a small fire. If, however, there is any chance that the fire may endanger others or may cause serious damage, confine or control the fire only if possible.

Use an appropriate extinguisher:

- Dry Powder (for all type of fire): Dry powder containing extinguishers are recommended for putting off all types of fire.
- CO₂ (for B and C type fire): Because the use of dry powder leaves a messy surrounding, CO₂ based extinguishers are often recommended for small fire of chemical origin.
- If fire breaks out close to a flammable gas supply or close to electrical power source, turn off gas supplies and electrical power sources.

Immediately after a fire extinguisher has been used, make a report of it and inform the Safety Officers.



If a solvent in a beaker catches fire, covering the beaker and depriving the fire of oxygen can easily extinguish the fire than using a fire extinguisher on the same beaker, which may cause the solvent to spill, thus increasing the hazard!

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If there are injured victims, provide the minimum necessary first aid '*Only If You Are Sure That There Is No Danger To Yourself*'. If providing assistance will endanger you, *DO NOT* attempt intervention and move the victim immediately to the nearest hospital. In case of urgency, call:

- 1. Ambulance 102
- 2. Fire station 101
- 3. Disaster management 108
- 4. A.P. State Emergency Service And Fire Station, Tirupati 0877 226 0101

The above list is not complete and there could be many other scenarios, not listed here. In such cases your response should be based on the given scenario.

If CHEMICAL EMERGENCY OCCURS:

Chemical emergencies such as large spills, spills involving highly hazardous or flammable materials, releases of toxic or corrosive gasses or substances should be treated as other types of emergencies. PULL THE FIRE ALARM AND EVACUATE THE BUILDING.

The above list is not complete and there could be many other scenarios, not listed here. In such cases your response should be based on the given scenario.

7. Institute Policies regarding Safety

7.1 Safety Audits / Inspections

Safety officers along with few students (on rotation) will visit each laboratory to ensure all the basic safety rules are in place. For chemistry laboratories, the following eight areas have been identified for observing and maintaining safety of highest standard:

1. Safe working practices 2. Safety equipment 3. Fire handling and evacuation process 4. Ventilation of chemical vapor 5. Safety in conducting experiments 6. Laboratory equipment 7. Storage and labeling 8. Disposal of liquid and solid waste.



7.2 Revision of Safety Manual

Safety Manual will be reviewed once in six months or whenever there is a necessity for a certain policy change.

A *quick run through* of lab safety rules/policies:

- Report "All" accidents, no matter how minor, to the Supervisor/Safety In-Charge immediately
- Do not work alone in the laboratory.
- Know the location of the (i) "Emergency Exits in the lab and instrument room and (ii) fire extinguishers.
- Student with medical/Health concerns should seek the advice of a Doctor before attending labs.
- Wear safety goggles and lab coat at all times. If you have spilled chemical in your eyes, flush with water in an eye wash station for 10 to 15 minutes. Use safety shower in case of chemical spillage on body. Notify the incident to Supervisor and Safety In-Charge.
- Always wear full sleeves and a lab coat while working in the lab
- Wear appropriate shoes while working in the lab. Feet must be adequately covered. Open toed shoes or sandals are not permitted in the laboratory.
- Confine long hair whenever working in the laboratory.
- NO tobacco products in the laboratory.
- Ensure safe handling of chemicals by referring to Material Safety Data Sheet (MSDS) or ask the supervisor
- Report all spills especially mercury spill to Supervisor and Safety in Charge.
- Segregate the waste solvents and solid wastes appropriately for proper disposal.
- Do not use broken or chipped glassware and dispose them in the glass disposal box.
- Used syringe needles should be dropped in syringe disposal box, and do not dump waste paper in the broken glass/needle disposal boxes.
- Do not perform unauthorized experiments in the lab.
- Avoid crowding in lab benches (not more than **6** in each work bench)
- Do not use earphones/headphones while working in lab
- Follow all the special instructions and be careful while handling & disposing bio hazardous samples.





SAFETY @ IISER TIRUPATI PHYSICS LAB

First Edition

Department of Physics IISER TIRUPATI



YOUR LIFE IS MORE IMPORTANT THAN YOUR 'DREAMS'!

IISER Tirupati is committed to ensuring the health and the safety of every individual working in the institute. We strive to make the necessary arrangements for everyone's safety within the institute. However, no safety measures are of use unless there is active cooperation of the individual under concern. Therefore, we urge everyone to follow the safety policies and regulations of the institute.

This safety manual is primarily targeted to the students, even though it is very well applicable to the teaching assistants, the technical assistants, and the faculty members of IISER Tirupati. Students include the BS-MS, i-Ph.D., and Ph.D. students and project fellows. Most of the work-related accidents occur to those who lack experience in handling devices of a potential safety hazard. Therefore, it is imperative that all the students and the staff of the institute understand the dangers related to the experiments in the labs and take proper care to avoid any accidents.



So, "look down the road" and have a safe life at IISER Tirupati!



Safety at IISER Tirupati [Department of Physics]

There are several safety regulations one should keep in mind in an academic-cumresearch institute. This booklet focuses on the safety measures the students working in Physics Laboratories – both teaching and research – must follow for a safe working atmosphere in our laboratories.

General safety guidelines for the Physics UG and Research Labs

- Before you start working in a teaching/research lab, carefully read the safety guidelines available in the lab and acknowledge that you understand the safety rules and regulations.
- Self-assessment of the safety of your working environment is perhaps the most important thing when it comes to being safe! If you notice that something does not look safe, report it to the lab technicians and the faculty members concerned with safety.
- Keep your laboratory clean and organized.
- Food or drinks (including water) are prohibited inside a teaching/research lab.
- Never work alone in a laboratory.
- Do not try to operate an instrument unless you know what you are doing.
- Wear shoes in a teaching/research lab especially when there are potential physical hazards (such as a sharp or heavy object that can fall on your foot).
- Experiments involving chemicals:
 - All chemicals (including water!) should be labeled clearly.
 - Never use a chemical from an unlabelled bottle.
 - Never smell or taste a chemical which is unknown (or known!) to you.
 - Avoid direct exposure to chemicals (wear dedicated gloves for handling chemicals).
 - Glassware used to treat dangerous chemicals must be cleaned very well after use.
 - Do not use bare hands to handle broken glassware. Use a dedicated glove.
 - Experiments involving volatile toxic chemicals must be performed inside a fume hood.
 - \circ Chemical waste should be disposed of in the containers dedicated to them.
 - Wash your hands thoroughly after working with chemicals (even if you used gloves).
- Experiments involving lasers:

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- The lasers in the Physics UG labs are relatively safe. However, they may pose a threat when used with focusing optics such as lenses. Wear safety goggles when working with lasers of class 3 or above.
- Avoid any circumstance where the laser is directed at you even by reflection especially when focusing optics are involved.
- When you are working with a laser, do not bend down such that you head is at the level of the laser.
- Experiments involving electrical hazard:
 - The mains supply in India is 230-240 V / 50 Hz AC. Shocks from the mains can be deadly. All the mains cables must be checked for any damage. They must be plugged in to the socket before switching on the electrical equipment. The equipment must be switched off before unplugging the cable. Never unplug a cable by pulling the cord.
 - AC voltages are more dangerous than an equivalent DC voltage because the body has much lower resistance to AC compared to DC.
 - Never touch bare cables/electrodes in an electrical circuit or power supply! They may carry lethal voltages.
 - Take great care while working with experiments involving high voltage/current. In some experiments, e.g., Arc emission spectrometer in UG lab, even after the power supply is switched off, the potential difference between the arc electrodes remain nearly the same for several minutes unless the electrodes are discharged. If the electrodes carry the high potential, it can pose a great threat to life.
 - All the electrical cables used in the experiments must be properly insulated. Presence of any faulty cable should be reported and replaced immediately.
- Experiments with potential physical hazard:
 - Some experiments require the use of sharp or heavy objects that can injure you seriously unless handled carefully. Make sure that you wear shoes while working on these experiments.
 - Some experiments involve heating objects (metal and oil) up to nearly 100
 °C. These can cause severe burns if not handled with care.
- Never leave the experimental setups unattended when they are operated with open flames, heated substances, live electrical cables, switched on lasers, etc.



- Spills in labs should be cleaned up immediately.
- After every experiment, tidy up the workspace and keep all the components at their respective places.
- Wash your hands thoroughly when you leave the lab after your lab hours.
- Twice in every month, scrutinize the safety measures in the lab and keep things organized.
- In case of fire inside a UG laboratory, and if the technical assistants or faculty are around, inform them about it and follow their advice. You may be advised to leave the lab immediately through the nearest exit, inform the security, and gather on the cricket ground.
- In a research laboratory, follow the instructions as per the general safety guidelines described in detail in the main Safety Manual of IISER Tirupati.
- Ensure unblocked access to the following:
 - Doors and windows
 - Fire extinguishers
 - Electrical panels

Helpline Numbers [General]

- Fire station: 101
- Ambulance: 102
- Blood Requirement: 104
- Disaster management: 108
- Gas leakage: 1906
- A.P. State Emergency Service and Fire Station, Tirupati 0877 226 0101



Major safety concerns in Undergraduate (UG) labs

Chemical Safety in UG labs:

One of the major safety concerns in a laboratory is handling chemicals. Even though there are no highly toxic chemicals in Physics UG labs, one should be aware of the potential health hazards in handling various chemicals used in the labs. Experiments which use various chemicals in Physics UG lab are the following:

S. No.	Chemical	Chemical Formula	Experiment	Dangerous? *
1	Glycerol	$C_3H_8O_3$	Viscosity measurements	No
2	Isopropanol	C ₃ H ₈ O	Several experiments	Yes
3	Potassium permanganate	KMnO₄	Arc Emission Spectrometer	Yes
4	Nitrobenzene	$C_6H_5NO_2$	Kerr Effect	Yes
5	Lithium Fluoride	LiF	X-Ray Diffraction	Yes
6	Bismuth Telluride	Bi ₂ Te ₃	Scanning Tunnelling Microscope	Yes
7	Manganese Sulphate	MnSO₄	Quincke's Method for magnetic susceptibility	Yes

* For details of the safety in using these chemicals, refer to the section <u>Chemical Safety</u> <u>Documentation</u>. Also, refer to <u>Globally Harmonized System (GHS) Pictograms</u>.



Electrical safety in UG labs:

There are dangerous high voltage sources in the Physics UG labs. The voltages can be up to 30 kV (for instance in XRD device, but this device is well-protected with interlocks so that you are completely safe to operate the machine as if there is no voltage at all). In the Kerr-effect apparatus, the voltage can go up to 10 kV, but the supply is configured to a maximum voltage of 3-4 kV. The maximum current the supply can provide is one milliamp which can indeed cause an unpleasant electric shock. The equipment with risky voltages/currents are:

S. No.	Experiment	Device	Maximum Voltage (V)	Maximum Current (A)
1	Kerr Effect	HV DC power supply	3.5k	0.001
2	Zeeman Effect	DC Power supply for Hg vapor lamp	30	10
3	Magnetic Susceptibility	DC Power supply for the electromagnet	35	3.5
Λ	Arc Emission	Power supply for arc electrodes	100	10
-	Spectrometer	Power supply for Mercury vapor lamp	4k	0.03
5	X-Ray Diffraction	Power supply for accelerating electrons	35k	0.001
6	Hall Effect	DC Power supply for the electromagnet	30	3.95
7	Biot-Savart's Law	DC power supply for the electromagnet	30	20
8	Hydrogen lamp	DC power supply	3.5k	0.01
9	Rectifier	Centre-tapped transformer (14-0-14)	28	1
10	General use	DC power supply	32	10
11	General use	DC power supply	30	2
12	General use	Transformer (AC)	9	0.5

For details of the danger associated with electrical safety, refer the section <u>Electrical</u> <u>Safety Documentation</u>.



Laser Safety in UG labs:

There are continuous wave lasers (diode lasers and diode pumped solid state lasers) in the UG lab. They belong to Class 3R (maximum power up to 5 mW). They are relatively safe. However, long direct exposure or exposure through a focusing optic like a lens can lead to eye injury.

S. No.	Experiment	Laser Type	Wavelength (nm)	Power (mW)	Class
1	Fabry-Perot Interferometer	Diode laser, continuous	650	5	3R
2	Kerr Effect	DPSS, continuous	532	5	3R
3	Waveplates	Diode laser, continuous	657.16	5	3R
4	Malus Law	Diode laser, continuous	650	3	3R
5	Ultrasonic diffraction	Diode laser, continuous	650	3	3R
6	Diffraction experiments	Diode laser, continuous	650, 532	3	3R
7	Michelson Interferometer	Diode laser, continuous	532	5	3R

For details about the dangers involved in working with lasers, refer to the section <u>Laser</u> <u>Safety Documentation</u>.



Major Safety Concerns in Research Labs

Electrical Safety in Research labs:

Lethal high voltage is one of the major life-threatening agents in our research labs. Look for the signs of high voltage in these labs and take proper care. Never touch an uninsulated electric cable in an electrical circuit without switching off the supplies. Below is a list of power supplies used in our research labs.

S. No.	Device	Model	Maximum Voltage (V/Vp)	Maximum Current (mA)	Lab Concerned
1	ISEG HV DC power supply	NHQ 205M	5000	2	
2	Stahl DC HV Power supply	HV 400-16	400	0.07	H
3	RF Output for Ion funnel		100	-	lesearc
4	RF Output for quadrupole ion guide		200	-	ch Lab
5	RF output for quadrupole mass filter		500	-	
6	RF output for ion trap		500	-	

For details of the danger associated with electrical safety, refer the section <u>Electrical Safety</u> <u>Documentation</u>.

Chemical Safety in Research labs:

S. No.	Chemical	Chemical Formula	Usage?
1	Isopropanol	C ₃ H ₈ O	General cleaning
2	Ethanol	C ₂ H ₅ OH	General cleaning
3	Methanol	CH ₃ OH	Preparation of samples for electrospray
4	Acetonitrile	CH ₃ CN	Preparation of samples for electrospray
5	Acetic Acid	CH ₃ COOH	Preparation of samples for electrospray

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Chemical Safety



Globally Harmonized System (GHS) Pictograms

Pictogram	GHS ID	What does It mean?	More information*
\bigotimes	GHS01 Explosive	 Unstable explosives Self-reactive substances and mixtures Organic Peroxides 	Explosive material
	GHS02 Flammable	 Flammable liquids, gases, solids Self-reactive substances and mixtures Pyrophoric substances Organic Peroxides 	<u>Flammability</u>
	GHS03 Oxidizing	 Oxidizing liquids, solids, gases 	Oxidizing agent
\Leftrightarrow	GHS04 Compressed gas	 Compressed Gases Liquefied Gases Refrigerated liquefied gases Dissolved Gases 	Compressed Gases
	GHS05 Corrosive	Corrosive to metals	Corrosive substance
	GHS06 Toxic	 Acute toxicity (oral, dermal, inhalation) 	Toxicity
(!)	GHS07 Harmful	 Acute toxicity Skin irritation Eye irritation Respiratory tract irritation 	<u>Lab Hazards &</u> <u>Risks</u>
	GHS08 Health hazard	 Carcinogenicity Reproductive toxicity Respiratory sensitization Germ cell mutagenicity 	<u>Carcinogen</u>

* 'More information' refers to the useful weblinks available in the e-copy of this document.



Chemicals used in UG and research labs

Chemical	Chemical Formula	Safe?	GHS Pictogram
Glycerol	C ₃ H ₈ O ₃	Safe	None
Isopropanol	C ₃ H ₈ O	Danger	
Ethanol	C_2H_5OH	Danger	
Methanol	СН₃ОН	Danger	
Acetonitrile	CH₃CN	Danger	
Acetic Acid	CH₃COOH	Danger	
Potassium permanganate	KMnO₄	Danger	
Nitrobenzene	$C_6H_5NO_2$	Danger	
Lithium Fluoride	LiF	Danger	
Bismuth Telluride	Bi ₂ Te ₃	Danger	
Manganese Sulphate	MnSO ₄	Danger	

Reference: https://chemicalsafety.com



Laser Safety



Lasers can be very dangerous! When?

Lasers can be of diverse types depending on their power output, the wavelength range of the light they emit (UV, visible or infrared), and the way the energy is delivered (continuous or pulsed).

The following symbol will be displayed wherever a laser is present.



Maximum Permissible Exposure (MPE): This term refers to the safe limit of the exposure of laser radiation to the eye. It is the maximum amount of power or energy density $(J/cm^2 \text{ or } W/cm^2)$ of a light source that is considered safe, i.e., the negligible probability of causing damage to the eye. The MPE is measured at the cornea of a human eye or the skin for a given wavelength and exposure time.

The classification of lasers based on the seriousness of the danger they pose is given below.



Source: www.lasersafetyfacts.com



The details of the classification of lasers are illustrated in the table below.

Class	Description	Warning label
1	 Safe under all conditions of normal use! 	
	 No precautions required E.g., A high-class laser with proper protection so that any danger is only when the protection is removed when the laser is running. 	CLASS 1 LASER PRODUCT
1M	 Safe except when operated with focusing optical elements The lasers emit large diameter or divergent beams 	LASER RADIATION DO NOT VIEW DIRECTLY WITH OPTICAL INSTRUMENTS CLASS 1M LASER PRODUCT
2	 Visible lasers (400-700 nm) with a maximum of 1 mW of continuous wave radiation Safe because blink reflex limits the eye exposure of the laser to less than 250 ms 	LASER RADIATION DO NOT STARE INTO BEAM CLASS 2 LASER PRODUCT
2M	 Safe if the beam is not viewed through optical instruments Blink reflex limits the eye exposure of the laser. This laser class also applies to larger-diameter or diverging laser beams. 	LASER RADIATION Do not stare into the beam or view Directly with optical instruments CLASS 2 M LASER PRODUCT
3R	 Safe if handled with restricted beam viewing Maximum permissible exposure (MPE) can be exceeded, but the injury is very likely minor Visible continuous wave lasers up to 5 mW fall under this category 	LASER RADIATION AVOID EXPOSURE TO BEAM CLASS 3R LASER PRODUCT
3B	 Hazardous to the eye if exposed directly Diffuse reflections are not harmful Use protective eyewear while operating the laser Laser safety signs lightboxes 	LASER RADIATION AVOID EXPOSURE TO BEAM CLASS 3B LASER PRODUCT

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	 should be used with lasers that require safety interlock They should be equipped with a key switch and a safety interlock 	
4	 Utmost care required while operating this type of lasers May cause damage to the skin and eye Even diffuse reflections can be dangerous They may also represent a fire risk They should be equipped with a key switch and a safety interlock 	LASE AVOID EY DIRECT O CLAS





Electrical Safety



Electricity can kill. Respect it!

Lethal voltages present in the labs can be identified by the following symbol.



Should you be concerned with high voltage or high current? In fact, it is a bit of both! If the voltage is not high enough, it may be relatively safe to touch an electrical circuit which can deliver high current. On the other hand, if the voltage is very high, but the current that the supply can deliver is very low, you might still be safe. Let us see why! The electricity is nothing but the flow of charged particles. In most of our everyday life situations, it is the flow of electrons. The electrons flow from a high potential to a lower potential. What happens when you get a shock is that the electricity flows through your body from an electrical circuit carrying a high potential to the ground.

Your body has certain electrical resistance. From the tip of your finger to your feet, it is about 100 k Ω under normal circumstances. However, if you are wet, the resistance can drop to about one kilo-ohm! Thus, if you are touching a 100 V terminal, the current that can flow through your body is about one milliamp under normal conditions. The resultant shock is barely perceptible (See the table below)! However, once the current passes through the body, its resistance decreases and more current starts flowing, which is indeed dangerous. If your body is wet, the initial current can be up to 100 mA, which is indeed fatal! However, if the power supply can deliver a maximum of one milliamp of current, and if it is not faulty, you are very likely safe! If there is some failure in the grounding of the power supply, this factor will not be applicable! Therefore, do not touch any electrical terminals that look suspicious.

One important thing to note at this point is that once the electricity starts flowing through your body, your resistance will decrease drastically facilitating more current to flow.

Given below in a box is a description of how human body responds to various amounts of currents [Source: Fish, R. M. & Geddes, L. A. Conduction of electrical current to and through the human body: a review. Eplasty 9, e44 (2009).]



1 mA	Barely perceptible
16 mA	Maximum current an average man can grasp and "let go"
20 mA	Paralysis of respiratory muscles
100 mA	Ventricular fibrillation threshold
2 Amps	Cardiac standstill and internal organ damage
15/20 Amps	Common fuse or breaker opens circuit*

*Contact with 20 milliamps of current can be fatal. As a frame of reference, a common household circuit breaker may be rated at 15, 20, or 30 amps.



Biosafety Manual

Department of Biology, IISER Tirupati

First Edition



Scope

This biosafety manual is relevant for all research activities that especially include handling biological material in the laboratories.

Introduction

The research personnel in biological laboratories are at risk of handling samples that may be pathogen infected or live organisms that pose a hazardous effect to humans as well as environment. Thus this manual will provide guidelines to adoption of appropriate methods to avoid hazards in microbiological laboratories, dangerous chemicals, liquid nitrogen etc.

Risk Assessment:

Before starting any research activity in the lab, risk assessment must be performed to identify the hazards and design possible control methods to prevent risk.

For risk assessment, the following steps are necessary:

A. Identification of risk related to cell culture related biological materials that includes pathogen and human or animal cells being handled

All material of human origin, including cell lines, tissue, and blood, must be considered potentially infectious for hepatitis and HIV and handled under Universal Precautions, which reasonably preclude cutaneous, oral, and parenteral exposure to personnel.

For risk identification of the pathogen, the researcher should be informed for the following:

- (i) Routes of transmission: The probable routes of transmission are
 - (a) Exposure of the agent through the skin, eyes and mucous membranes.
 - (b) Piercing by a syringe needle or other contaminated sharp.
 - (c) Bites, scratches from infected animals.
 - (d) Ingestion of liquid suspension of an infectious agent.



(e) By contact of the contaminated hand to mouth exposure.

(f) Inhalation of infectious aerosols

(ii) Infective dose

(iii) Stability in the environment

(iv) Host range and its endemic nature.

(v) Method of treatment available.

B. Identification of risk related to procedures carried in the laboratory

Safety procedures must be established for the following laboratory handling procedures:

(i) that generate aerosols as they are invisible and spread faster.

(ii) on animals as they can transmit pathogen through saliva, urine and faeces.

(iii) Genetic manipulation of microorganisms can lead to generation of higher virulent microorganisms or antibiotic resistance.

C. Classify the microorganisms into appropriate biosafety levels

Based on the above information, the laboratory can be assigned at various levels that would help in the containment of the pathogens/human/animal cells. The containment is achieved by defining the safety of the research by various biosafety levels. Overall following biosafety levels are followed:

Biosafety Level 1 (BSL1)

BSL1 is defined for experiments that include a pathogen that is unlikely to cause any disease in humans or animals. Thus special facility or equipments are not needed but still the laboratory personnel should following initial training for carrying out experiments in BSL1. Following are the standard practices for BSL1



- 1. Lab coats, eye protection and toe-covered footwear to prevent contamination must be used.
- 2. Surgical masks should be used for handling carcinogens, chemicals or for biological agents if there is a risk of spread of infection through inhalation.
- 3. Protected glasses should be used during UV transilluminators.
- 4. Face shields must be used to protect the personnel from splashes.
- 5. Storage of food items or eating is not allowed in the laboratory
- 6. Mouth pipetting should not be practiced but pipetting devices should be used.
- 7. Broken glassware should be handled with dustpan and brush
- 8. In case of spills, contain the area and decontaminate the area using recommendations for that spill.
- 9. Waste material should be discarded in designated waste disposals.
- 10. The lab in charge should provide initial training for lab personnel about potential hazards and precautions to be taken and they are updated whenever there are changes in procedures.

Biosafety Level 2 (BSL2)

BSL2 is defined for experiments that include a pathogen that could cause disease in humans or animals but is unlikely a serious hazard. Also, effective treatment and preventive measures are available and the risk of spread of infection is limited. Thus BSL2 is a contained area where lab personnel requires specific training and are supervised by competent researcher.

- ⇒ Therefore, the lab doors must be self-closing type and there must be provision for locking the same.
- ⇒ The international biohazard-warning symbol must be displayed on the doors of the labs where microorganisms of Risk Group 2 or higher risk groups are handled. Name of the PI and contact details must also be displayed.
- ⇒ The pathogens must strictly be handled with biosafety cabinets BSLA2 type (described below). Also, the biosafety cabinet must be located away from doors and windows that could be opened which could create disruptions in the airflow.
- \Rightarrow Liquid disinfectant trap must be provided to vacuum lines.
- \Rightarrow The access to the laboratory is restricted to trained researchers.
- \Rightarrow Medical surveillance and immunization must be provided to the lab personnel.
- \Rightarrow Spills involving contaminated material must be decontaminated and cleaned up.





Biosafety Level 3 (BSL3)

BSL3 is defined for experiments that include a pathogen that has a potential to cause serious human or animal disease and is lethal through exposure by inhalation. Effective treatment and preventive measures are available

- ⇒ BSL3 is a strictly contained facility with special engineering and design features. Access is restricted and through two self-closing doors with a change room in between. The space between exit and window should be capable for sealing or fumigation.
- ⇒ Vaccum lines must be protected with high efficiency particulate air (HEPA) filters. The ventilation system must provide directional airflow by drawing air from clean areas to contaminated areas.
- ⇒ The decision to implement Biosafety Level 3 recommendations in the laboratory may only be made by the Institutional Biosafety Committee.
- ⇒ Entry into a BSL-3 facility is restricted to those individuals who have taken training and have demonstrated knowledge of BSL-3 Standard Operating Procedures and Safety Practices by means of a written exam.
- ⇒ All experiments involving the manipulation of infectious material are carried out in biological safety cabinets (BSCII or III) or other physical containment devices.
- ⇒ The personnel are required to wear appropriate personal protective clothing and devices to protect themselves from the exposure to pathogen. The personnel



should be wrapped around in gowns or coveralls. The clothing needs to be decontaminated before discarding.

 \Rightarrow Eye and face protections should also be decontaminated.

Biosafety Level 4 (BSL4)

BSL4 is defined for experiments that include a pathogen that usually causes serious human or animal disease and that can be readily transmitted from one individual to another, directly or indirectly. Effective treatment and preventive measures are not usually available.

Since it is life threatening with no effective treatment, additional measures are followed for BSL4.

- \Rightarrow Access to the laboratory is strictly restricted and is through an airtight doors.
- ⇒ A shower is provided to decontaminate the surface of the pressure suit before the person leaves the work area.
- \Rightarrow The exhaust air from the suit area must be filtered by two sets of HEPA filters.
- ⇒ Double door autoclave for decontamination of disposable waste materials from the suit area to be used.

Summary of Classification of Biological Agents According to Risk

(Modified from Biosafety in Microbiological and Biomedical Laboratories, Centers for Disease Control and the National Institutes of Health, 3rd Ed., 1993.)

Biosafety Level 2: Bacterial Agents†

Bacillus anthracis

Bordetella pertussis

Campylobacter - all species

Chlamydia psittaci, C. Pneumoniae, C. trachomatis

Clostridium botulinum, C. tetani

Corynebacterium diptheriae



Leptospira interrogans- all serovars

Legionella pneumophila; other Legionella-like agents

Mycobacteria except M. bovis or M. leprae

Neisseria gonorrhoeae, N. Meningitidis

Pseudomonas psudomallei

Salmonella - all serotypes

Shigella - all species and all serotypes

Vibrionic enteritis (Vibrio cholerae, V. parahaemolyticus)

[†] Additional primary containment and precautions, such as those described for work at the BSL-3 level are recommended for activities with high potential for droplet or aerosol production, for work with antibiotic - resistant strains and for activities involving production quantities or concentrations of infectious materials.

Biosafety Level 2: Fungal Agents

Blastomyces dermatitidis

Cryptococcus neoformans

Sporothrix schenckii

Pathogenic members of the genera Epidermophyton, Microsporum and Trichophyton

Miscellaneous molds

Cladosporium (Xylohypha) trichoides

Cladosporium bantianum

Penicillium marnefii

Exophiala (Wangiella) dermatitidis



Fonsecaea pedrosoi

Dactylaria gallopava (Ochroconis gallopavum)

Biosafety Level 2: Parasitic Agents

Nematode parasites of humans

Ascaris spp.

Stronglyoides spp.

Hookworms Enterobius spp.

Protozoal Parasites of Humans

Toxoplasma spp.

Plasmodium spp.

Trypanosoma spp.

Entamoeba spp.

Coccidia spp.

Giardia spp.

Leishmania spp.

Sarcocystis spp.

Cryptosporidia spp.

Trematode Parasites of Humans

Schistosoma spp.

Fasciola spp.

Cestode Parasites of Humans



Echinococcus granulosus

Taenia solium (cysticercus cellulosae)

Hymenolepsis nana.

Biosafety Level 2: Viral Agents ††

Hepatitis A, B, C, D, and E virus Herpes viruses - except Herpesvirus simiae (Monkey B virus) which is BSL-4

Influenza virus

Polioviruses Poxviruses - all types except variola which is restricted Rabies virus

Mouse hepatitis virus. For animal work it needs a separate contaminant suite.

†† Additional primary containment and personnel precautions, such as those described for Biosafety Level 3, may be indicated for activities with potential for droplet or aerosol production and for activities involving production quantities or concentrations of infectious materials.

Biosafety Level 3: Bacterial Agents

Brucella - all species

Francisella tularensis

Mycobacterium bovis; M. tuberculosis Yersinia pestis

Biosafety Level 3: Fungal Agents

Coccidioides immitis

Histoplasma capsulatum

Biosafety Level 3: Viral and Rickettsial Agents

Lymphocytic choriomeningitis virus (LCM)



Rickettsiae - all species when used for transmission or animal inoculation experiments Vesicular Stomatitis Virus

Retroviruses, including Human and Simian Immunodeficiency viruses (HIV and SIV) Transmissible Spongiform Encephalopathies (Creutzfeldt-Jakob, kuru and related agents)

Arboviruses and Arenaviruses

Biosafety cabinets

Biosafety cabinets are one of the essential requirements for maintenance as well as containment of pathogen in a biosafety facility.

The selections of BSCs are decided on the type of protection needed. This includes

Lab personnel protection

- Sample protection
- Risk involved with the pathogen
- Risk involved with the chemicals

Following are the guidelines for biosafety cabinets to be considered for different biosafety levels;

Class I Biosafety cabinet

The Class I BSC protects the personnel and the environment from exposure to biohazards, however samples are vulnerable from coming in contact with airborne contaminant present in the room. Thus they are used for general microbiological research with low- and moderate-risk agents. The disadvantage is that it does not provide protection from contamination of the sample that is being handled as the room air passes over it.

Class II Biosafety cabinet

The Class II BSC provides protection both to personnel as well as the sample. These cabinets are suitable for BSL2, 3 and 4. The cabinet has an open front with a stream of inward air preventing the escape of aerosols outside. The air flowing downward over the



working surface is HEPA filtered and thus the cabinet is free of airborne contaminant protecting the sample from contamination.

Class III Biosafety cabinet

BSC III cabinets provide an absolute safety where the cabinets are of welded metals construction and are gas tight. It has a non-opening window and the work is carried out glove ports in front of the cabinet. Thus they are working with highly infectious pathogenic agents and provide maximum protection for the environment and worker.

Below are the details of different types of BSC cabinet suitable for different biosafety levels:

Inflow Class Velocity		Recycle Air	Exhaust	Control Plenum	Exhaust	Biosafety	
	(m/s)	(%)	Air (%)	Surrounded by	Alternatives	Level	
Ι	US:0.38 EN:0.70	0	100	Outside Air	Inside room / Hard duct	1, 2 & 3	
ll Type A1	US:0.38 EN:0.40	70	30	Outside Air	Inside room / Thimble duct	1, 2 & 3	
ll Type A2	US:0.50 EN:0.40	70	30	Negative Plenum	Inside room / Thimble duct	1, 2 & 3	
ll Type B1	US:0.50 EN:0.40	30	70	Negative Plenum	Hard duct only	1, 2 & 3	
ll Type B2	US:0.50 EN:NA	0	100	Negative Plenum	Hard duct only	1, 2 & 3	
ш	Closed: * >0.5"WC	0	100	Negative Plenum	Inside room / Hard duct	1, 2, 3 & 4	

Source: Laboratory Biosafety Manual. Second Edition (revised). Interim guidelines. World Health Organization. p32.

* Pressure differential between chamber and environment.

Emergency responses

In case of spill inside biosafety cabinet

- 1. Contain aerosols by leaving BSC cabinet operating
- 2. Disinfect the cabinet by treating with 10% Bleach.
- 3. Clean the cabinet with water followed by 70% ethanol.
- 4. Operate the cabinet for 15min before starting work.

In case of spill outside biosafety cabinet

If the spill is of BSL1 or 2



- 1. Remove the contaminated clothing or gloves. Wear fresh gloves and If possible contain the spill by covering with paper towels. Decontaminate the area by addition of 10% bleach at least for 20min and mark the area of spillage.
- 2. Notify other lab personnel for spills.
- 3. Clean up the bleach
- 4. Dispose off all the materials in biohazard waste.

If the spill is of BSL3

- 1. Notify other lab personnel for spills.
- 2. Remove the contaminated clothing or gloves.
- 3. Wash body parts with disinfectant soap
- 4. Notify the emergency response team.

Biohazard Waste

Non-contaminated waste can be treated as non-infected waste and can be disposed off as general waste. The following waste can be treated as potentially biohazard waste and should be segregated and disposed off based on the waste type in a responsible manner.

Sharps Waste

All needles, syringes (with or without the attached needle), Pasteur pipettes, scalpel blades are collected in a puncture proof sharp containers for disposal.

Microbiological Waste

All materials containing or in contact with cultures of microbiological organisms, patient specimens sent for microbiological culture or items contaminated by patient specimens are collected in autoclavable red bags with a biohazard sign. The materials are then autoclaved for decontamination and picked by the specified agency of the institute for incineration.

Biological Materials Waste

All discarded vaccines, immunoglobulin, plasma, albumin, blood or tissue fractionation products, enzyme preparations. Similar procedures are followed as for microbiological waste.



If the waste is liquid, then it is treated with 10% bleach solution overnight before discarding.

Animal Pathogen Contaminated Waste

All bedding and other materials contaminated with blood, excreta or secretions of animals infected with transmissible human or animal pathogens. Similar procedures are followed as for microbiological waste.

General Laboratory safety guidelines:

- Wear approved lab coat at all the times. If you have spilled chemical in your eyes, flush with water in an eye wash station for 10 to 15 minutes. Use safety shower in case of chemical spillage on body. Notify the incident to Supervisor and Safety In-Charge.
- Confine long hair whenever working in the laboratory.
- Always wear full sleeves and a lab coat while working in the lab
- Wear Shoes while working in the lab. Feet must be adequately covered. Open toed shoes or sandals are not permitted in the laboratory.
- NO tobacco products in the laboratory.
- Food items and Eating is NOT allowed in the laboratory.
- Familiarize yourself with the lab (equipment, chemicals).
- Never mouth a pipette, use a rubber bulb.
- Report all spills to Supervisor and Safety in Charge.
- **Do not use broken or chipped glassware** and dispose them in the glass disposal box.
- There is a separate disposal for plasticware contaminated with organic solvents. Thus make sure that contaminated plasticware are discarded in the specified container present in each lab.
- Do not discard organic solvents in the sink but in the waste container for organic solvent placed in each lab.