

Experimental Biotechnology
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Module-I
Basics of Laboratory Research
Lecture-01
Good Lab Practices (Part-1)

(Video Starts: 00:23) (Video Ends: 00:59) Hello everybody this is Dr. Vishal Trivedi from Department of Biosciences and Bioengineering IIT Guwahati and the course experimental biotechnology is more about the experiment what you are going to perform in your laboratory or what you are going to perform in your laboratory. So, before you enter into any laboratory, you have to follow the certain rules and regulations, and as well as you have to follow the guidelines.

Just like when you start learning the motorcar or any scooter and you want to run it on the road, you have to follow the rules of the road and you have to follow all the traffic signals, you have to understand what are those things and similarly, when you enter into an experimental lab whether it is the lab for your school lab or whether it is a lab for the research purposes. You have to follow the certain guidelines and all these guidelines are coming under the purview of good lab practices.

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Good Lab Practices (GLP)

An internationally recognized definition of GLP goes like this: Good Laboratory Practice (GLP) embodies a set of principles that provides a framework within which laboratory studies are planned, performed, monitored, recorded, reported, and archived.

New Zealand and Denmark were the first to introduce GLP in 1972. United States was the next to introduce GLP in response to the poor scientific practices prevalent in US around that time.

Why ???

An international economic organization, the Organisation for Economic Co-operation and Development (OECD) published the principles of GLP in 1981 under the name 'OECD Guidelines for the Testing of Chemicals' that were internationally accepted.



So, the good lab practices are the set of guidelines, what a person has to follow when he will going to perform the experiments in a laboratory. And as per the definition is concerned, the good lab practices are the practices which are actually embodied a set of principle that provides a framework within which the laboratory studies are planned, performed, monitored, recorded, reported and archived, which means the good lab practices are the set of the guidelines, which actually are going to follow by everybody when you are going to plan the experiments.

What are the different information you require, what are the different biohazards you are going to generate and what are the outcomes of that particular experiment. So, it is not important that you are doing an experiment. It is also important that you should do the experiment under the certain guidelines so that the results what you are going to come out from the experiments are actually be reproducible.

And the documentation is also going to be in a perfect shape so that the other people also could be able to follow the experiments, could be able to replicate in their labs and that is all under comes under the purview of the good lab practices, the question comes, why there is a need for

good lab practices. So, in the beginning of the 19 th centuries, the people were not following a good lab practices.

And as a result, they were actually not documenting the procedures, they are not documenting the data, they were not documenting the results properly. And because the scientific community wants to replicate those experiments, it was very difficult to replicate because the minute experimental details were missing. In some cases, it was very, very problematic because you can imagine that there is a biopharma company who is actually producing medicine or who is developing medicine.

This medicine could be for the cure of cancer and all other kinds of thing. And they are claiming something, like they are claiming that this particular medicine is going to cure the cancer patients, but the data what they are providing cannot be verified until they are not going to tell you the procedure. So, realizing this fact the food and drug administration of the United States of America, realize that people are doing the malpractices.

People are not reporting the total procedures and as a result they said that we actually require a set of guidelines, and that these guidelines have to be followed by all over the world. So, that the scientific community are going to perform the experiments with a set of rules and regulations and as a result, you can be able to perform these experiment, you can be able to reproduce these results.

And that is how it is actually going to bring more and more transparency. So, for this purpose the food and drug administration's are actually formulated the international economic organization, the organization for Economic Cooperation and Development that is OECD and the OECD is the organization which publishes the principles of GLP in 1981 under the name OECD guidelines for the testing of chemicals that were internationally recognized.

I have given you the link, which you can actually follow to understand and read the complete guidelines about this particular the OECD guidelines, what you have to follow. I am only going to give you a summary of some of these guidelines what you should follow when you enter into a

research lab or to the your school labs. So, that you should be able to protect yourself while you are doing these experiments.

On the other hand, whatever the data you generate could be based on the scientific principles, that could be reproduced by the other scientific community and it should at the end going to help the human society.

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Guide lines and Precautions

Recommended action

- Always wear the appropriate equipment of personal protection. They include an apron, a pair of gloves, safety glasses, and shoes.
- Read thoroughly the detailed procedure before carrying out the experiment.
- Before starting the experiment, label your materials (tubes, vials, specimens, etc.).
- Check the label on the container before taking out any substance.

Gloves Apron Safety glasses Shield Radioactive shield Radioactive Batch

So, there are guidelines which are recommended that you should follow this, and there are guidelines that you that says that you should not do this when you are inside the lab. So let us see what are the recommended actions. So, in the recommended actions, when you enter into a lab you always wear the appropriate equipment of the personnel protections. They include an apron, a pair of gloves, safety glasses and the shoes.

So, what you see is that in the lab you actually uses the 2 different types of gloves. One is called as the latex gloves. These latex gloves are only to protect or to protect you from the getting exposure to the chemicals or to getting the exposure to the some kind of dust and all of that kinds of material what you are generating in your laboratory. Apart from that these are the rubber gloves, which people normally use when they are actually handling the hot liquids.

Like, suppose you are boiling a solution into the microwave and you want to take out. So, these are their microwave gloves. Apart from that you always should wear the aprons, because the purpose of wearing the apron is that, in case there will be any spillage from the chemical reactions or if there would be any spillage from the . For example, if you are growing a bacteria which is infectious and if you are getting some spillage from the flask.

That spillage is not going to get into your own cloth. So, wearing an apron is actually going to protect you from getting the exposure to the infectious or these organisms, as well as getting the exposure to be direct exposure of the chemicals. So, if you wear the apron, apron is made up of thick cotton cloth. So, it actually kind of a first layer of protections that, you should not get directly exposed to the chemical as well as the infectious organisms.

Apart from that, you also have to wear the safety glasses, the safety glasses are mostly been used in chemistry laboratories where it actually protects your eyes because when you are doing some reactions and you do not know what kind of byproducts are going to be generated or what other products are going to be generated. So, when you are doing a reaction which is unknown or which is a newly you know you are doing something so in that cases, you have to protect your eyes.

So, you can use safety glasses and these safety glasses are actually going to protect you. Apart from that, you can also use the shield, the shield is actually a kind of a, you know, face shield, so that face shield is actually also going to protect you from the getting, you know, any kind of spillage onto the face. For example if these face shields are also been used very routinely in the hospitals or other kinds of experimental labs where you expect a spillage of the chemicals or the biological waste.

So, in those cases, it actually will not go into your face and it is actually going to protect your body. Apart from that, if you are working with a radioactive material, then you also should use the radioactive shields. So, in the radioactivity when you are working on a radioactive chemicals like if you are working with the TCM or you are working with the P 32 labeled ATP. Then you have to use the lead wall.

So, in the lead wall what you have. You have actually having the bricks, which are made up of brick lead bricks and these bricks has to be kept in front of you as a wall and then you are going to perform the experiment across the wall. So, the idea is that the any kind of radiation what is coming from the radioactive material is going to be, you know, going to be stopped or going to be blocked by the, these lead bricks.

And that is how it actually it is not going to expose your body. Apart from that, you also have to wear with these radioactive batches. The purpose of these radioactive batches is that when you build these, they actually are going to count how much radiation you are getting. So, when you are actually working with the radioactive material, especially like P 32 or the TCM. The beta particles what is coming from these materials are actually going to be a sense by these batches.

And then they are actually going to keep a count how much radiation you are getting, because it is not recommended that you get exposed too much to these radiation. So, in a timely manner you have to send these patches to the some of the radioactive agencies and what they will do is, they are going to count how much is the radioactive count present in that particular batch.

And if that count is beyond a limit, then they will ask you to remain away from the lab or at least not to perform the any experiment with the radioactivity because then actually it is going to be the life threatening. So, to monitor the health of the person who is performing the experiment with the radioactivity. You always have to wear these radioactive batches, so that where you are performing the experiments.

See because you know most of these radioactive radiations you cannot be able to see. So that is why these batches are actually going to tell you that how much your body is receiving the radioactive radiations and once it is crossing a particular limit, then they will ask you to wait and rest. Because once you receive the radiations the cells are going to die off, and then actually your radioactivity count is going to go down.

So, then you can wait for some time and then you are ready to work again. But during this information, only you will get when you hear these radioactive batches. Before you plan the experiment you have to read the procedures very thoroughly. So, that you will be able to know at what procedure you require and what are the equipments are required. Then, before starting the experiment, you have to label your material.

For example if you are using the or beakers, all these material has to be labeled so that there will be no mismatch. There will be no, like, you know, that you will not label the things and then you forget and then you are actually going to report that I have added x material, but instead you have added actually the y material. So, when the people will try to reproduce or we will try to replicate.

They will not be able to replicate and your documentation is also going to be very, very compromised. After that, you have to check the label of the container before using that container from the success, because if the material is hygroscopic you have to maintain the low moisture. So, you have to use it very carefully and then close the container. If it is a hazardous material or if it is a carcinogenic material.

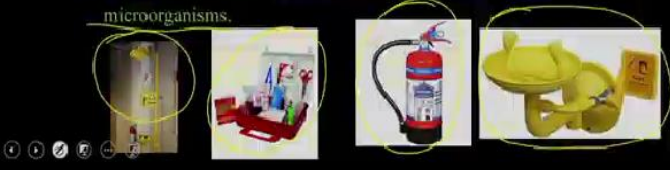
Then also you have to take the proper precautions, you have to wear the gloves, you have to take the full precautions or if it is a material which evaporates, like. So in those cases, you have to take the appropriate precautions.

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Guide lines and Precautions

Recommended action

- Be well aware of the locations and the operating procedures of the safety equipments such as safety shower, first aid kit, fire extinguisher, eyewash, etc.
- Use a chemical fume hood for strong acids and other fuming chemicals and solvents.
- Use the recommended biosafety level containment while handling microorganisms.



The image contains four small photographs arranged horizontally. From left to right: 1. A safety shower, which is a large white wall-mounted unit with a yellow emergency stop button. 2. A first aid kit, a red plastic box containing various medical supplies like bandages and antiseptics. 3. A fire extinguisher, a red cylindrical device with a black handle and hose. 4. An eyewash station, a yellow plastic basin with two eye-cupping nozzles and a yellow emergency stop button.

Apart from that, you also have to be aware of the location and the operating procedure of the safety equipments. So, you are going to use the safety showers, first aid kits, fire extinguishers and the eye wash. Before you start the experiment, before you enter into a laboratory you also should know, where are the safety equipments are being kept in the department. So, that whenever there will be any such accident occurs, you should know where I should go.

And use the particular type of equipments, what are the equipment you have, you have the safety showers, so safety showers are like the normal shower what you have in your home. And the purpose of the safety shower is only to wash away the chemicals what has been spilled onto your body or suppose some time you all got a small fire hazard or something you know your clothes have caught a fire.

In those cases, we need the safety shower and it is actually going to remove the chemicals. Apart from that, you can also have the first aid kits, in case you have got some minor injury or minor accidents, then you can have different types of bandages and all those kinds of things. So, that also is required when you are working in a laboratory. So, that in case there is a minor accident or minor injury, you can be able to take care of that.

Apart from that you also require the fire extinguishers. So, that actually is and required to, you know, to extinguish the fires. And it is not important that you should know the location of those

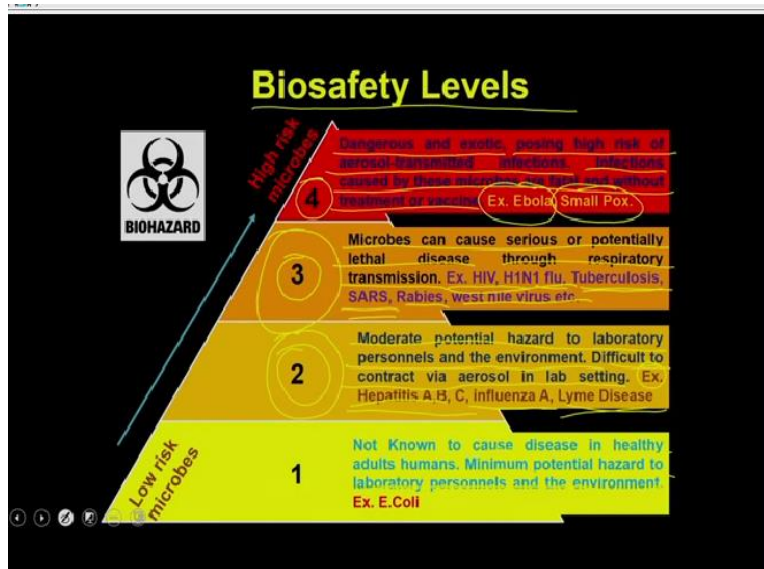
equipments, you also should know how to operate these equipments because when there is a fire you might not have the professional people to help you, in that case you might have to operate these equipment yourself.

And apart from that you also require the eyewash. So, eye wash is a equipment which actually you can use in case some there is a spillage of the chemicals in your eyes. In that case, you cannot use the showers, you can easily use the eye washes to wash your eyes and that actually is going to remove the level of chemicals in your eyes and probably can help you in terms of getting the recovery, very soon.

You have to use a chemical hook for strong acids and under the other fuming chemicals and solvents. So, for most of the chemistry labs what they are using is they are using very strong acid and strong basis or some of the chemical which are actually evaporated. So, like organic solvents and all those kinds of. So, all that you have to use a fume hood, because the fume hood is actually going to suck all these fumes and that is how they are actually not the person who is using them will not get exposed.

Apart from that, when you are working in the biology lab, you are going to use recommended biosafety containment while handling the microorganisms.

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So, let us understand what are the different biosafety levels. So, biosafety levels, according to the biosafety level you have to handle a particular microorganisms. There are 4 biosafety levels in a particular laboratory and the biosafety levels are being classified based on the low risk microorganisms versus the high risk microorganisms. So, you have the BSL 1 BSL 2 BSL 3 and BSL 4. Let us understand what is mean by BSL 1.

So, the BSL 1 category is actually are those organism, which does not cause any disease in healthy individuals. Minimum potential hazard to the laboratory personnel and the environment. The classical example is the E. coli, which means in the BSL 1 facility or BSL 1, the microorganism which falls under the BSL 1 does not cause any disease to a healthy individual. If you are immunocompromised or if you are having any kind of other diseases.

Then probably these microorganism may cause the disease, but for a healthy individual these microorganism will not cause any disease one of the classical example is E. Coli . Then you have the BSL 2, in the BSL 2 you have the moderate potential hazard to the laboratory personnel and the environment. Difficult to contact via aerosol in the lab setting. One of the classical example is the hepatitis A, B, C, influencer and the Lyme disease.

So, the BSL 2 organisms are also very moderate in some cases they may cause the disease but these diseases are not life threatening. This disease could be cured, simply by giving some medicines or they are actually manageable. The major thing, which is actually not the contaminant. So, the BSL 2 organisms will not get contaminate you buy the forming the aerosol, which means they are not airborne organisms/

You will get in exposure only if there will be a spillage and you will get a direct contact to these microorganisms, classical example is the hepatitis, the influenza, as well as the Lyme disease. Then you have the BSL 3, the BSL 3 organisms are the organism which can cause serious or potential lethal diseases, through respiratory transmissions. Examples are HIV, H1N1 flu, tuberculosis, SARS, rabies, west Nile viruses.

So, the BSL 3 facility organisms are the organism which actually can cause very serious disease or life threatening diseases. They also get a spread through aerosol, so that is why they are very, very infectious and they do have to protect yourself and from these micro organisms, the classical examples are the HIV H1 flu, tuberculosis, SARS and that is why have to use the appropriate biosafety cabinets or the other kinds of equipments to protect yourself.

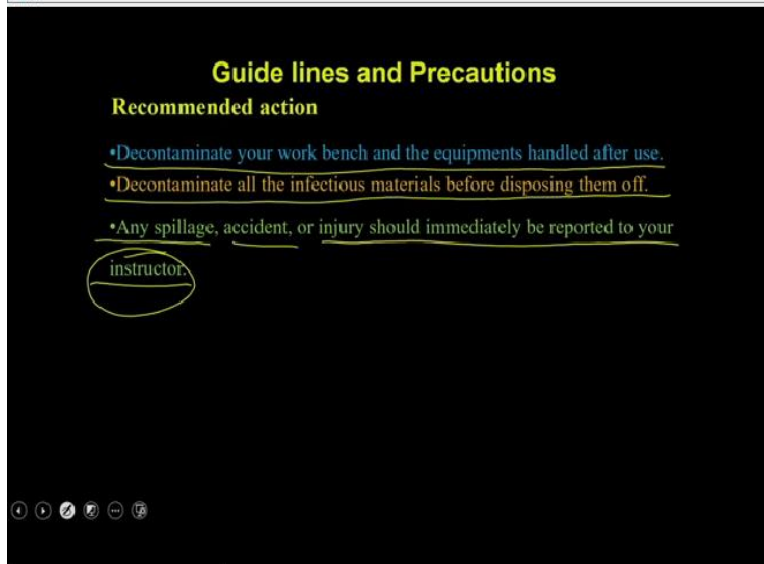
Then you have the BSL 4, in the BSL 4 you have the dangerous and the exotic posing high risk of the aerosol transmitted infections, infections caused by these micro organisms are fatal. And without treatment or the vaccine one of the classical example is the Ebola or the smallpox. So, BSL 4 is the category of the microorganism where, which actually cause the fatal diseases, and they are actually are life threatening.

So, in this case is the one of the classical example is the Ebola or the smallpox. So, all these biosafety levels are actually been used to classify the different microorganisms. So, that you will be able to buy the particular type of equipment, while you are handling these microorganisms. If you are using the BSL 1 and BSL 2, you can be able to use them or monitor them or manipulate them within the laminar hood or the biosafety cabinets.

Whereas for the BSL 3 and BSL 4 you require the, not only the biosafety cabinet but you also require a negative pressure room, so that you will be able to manage these microorganism, not infecting the personnel who is using that. And what is mean by the negative pressure is that you are actually going to suck the air from the room and then you are actually going to burn these air. So, that whatever the material comes out from these rooms are actually going to be burn at a very high temperature.

And that is how you are actually going to destroy all the microorganism or the aerosol what is being generated within these rooms.

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Apart from that, you have to decontaminate your workbench and the equipment handled after use, which means once you are done with the experiment suppose you are using the bacteria or other things, then you should decontaminate your pipettes, you should decontaminate your hands and you can decontaminate your workbench as well. Decontaminate all the infectious materials before disposing them off.

So, before you throw the bacteria or viruses or any other thing you have to ensure that they are being disposed, they are been sterilized. And so that they will not cause any potential hazard to the environment. In any case, when you have any spillage accident or the injury, you should directly report that to your lab supervisor or to the lab instructors, because it is important that the person who is more qualified.

And the person who have handled such situation should be informed so that he knows, how to cover up, how to take care of the spillage, because if you have done the radioactive spillage the procedure is different. If you have done the acid spillage, that is different and so on. So, the procedure is required a proper protocol has to be followed if you have done the radioactive spillage or any other kind of spillages.

But that is why it is important that if you have any such accident like fire accident, spillage, all that has to be reported to your instructor or to the supervisor.

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Guide lines and Precautions

Recommended action

Decontaminate all the infectious materials before disposing them off.

Sanitisation	Disinfection	Sterilisation
Surfaces cleaned and microbial load reduced	Microbes on surface killed, inactivated and made harmless	Total removal of any contamination - no viable microbes remain
The common solution for routine cleaning.	The solution for simple cases of contamination.	Reserved for serious contaminations.
A mild cleaning agent such as soap or household detergent will clean greasy surfaces but may have little antimicrobial activity.	Use quaternary ammonium compounds which have antimicrobial properties. They do not corrode metal or damage plastics.	Use oxidative agents such as bleaches or hydrogen peroxide. Stick to the safety rules and don't use these agents on metal or plastic.

Any spillage, accident, or injury should immediately be reported to your instructor.

And as I said you know decontaminate all the infectious material before you dispose them off into the proper dust bins or the trash can, you can have the 3 step of sanitization, you can have the sanitization, you can have disinfection and then you can have the sterilizations. In the sanitization you are going to do the surface clean, and you are going to reduce the microbial load.

That you can do simply by the common solution for routine cleaning. For example that you can use a mild cleaning agents such as the soap or the household detergent and that actually is going to sanitize the material or sanitize the surfaces. And that is good enough for reducing the load of the microbial load and you can add some material which also may have the little anti microbial activity.

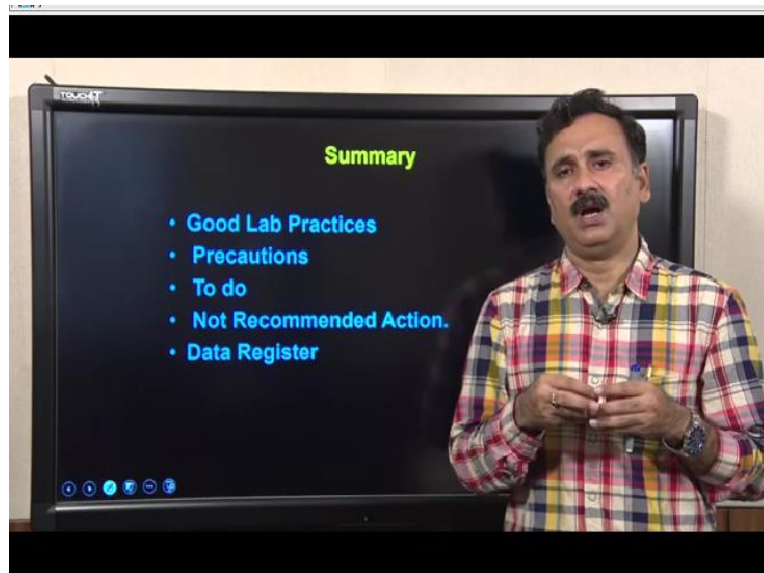
Then you can do a disinfection, disinfection means you are going to kill the microorganism, so the microorganism on the surface are being killed inactivated and made harmless, the solution for the simple cases of contaminations. You can use the quaternary ammonium compounds which are antimicrobial properties, they do not corrode the metal or the damaged plastics and then you have the third step, which is actually the sterilizations.

Sterilization means the total removal of any contamination, no viable microbes remain. This is reserved for the serious contaminations, you can use the oxidizing agents such as bleach or the hydrogen peroxide, stick to the safety rules and do not use these agent on the plastic or to the metal surfaces. So, you have the 3 different options depending on what kind of the spillage you have done.

If you can do sanitization, you can do disinfection, you can do the sterilization, because if the microbes what you are using is falling into the category of BSL 1 and 2, you can do sanitization and disinfection. But if you are working with the microorganism which is falling under the category of BSL number 3 and 4, then you have to do the sterilization. So, that the micro organism what you have spilled should be get killed.

And it should be removed from the surfaces, otherwise it because it is the airborne and it can actually cause the fatal diseases, it will be problematic for the person who is going to get exposed. And as I said, you have to report all the spillages accident and injury to your instructor or to the lab supervisors.

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With this I would like to conclude my lecture here. Thank you.