

TRANSCRIPT

Welcome students. Program Bachelor of Science. Third year subject Botany semesterr 5 paper code BOC 107 paper title microbiology and plant pathology. Title of the unit. Introduction to microbiology module name physical methods of sterilization. Model number two. The module consists of methods of sterilization in which we will study about physical methods of sterilization. Coming to the learning outcomes. At the end of this module the learner will be able to Explain the methods of sterilization, Differentiate between physical and chemical methods of sterilization. Explains the different type of physical methods of sterilization, sites example of instrument used in physical method of sterilization. Now coming to the methods of sterilization, there are two methods of sterilization which are physical methods and chemical methods. Now sterilization by physical methods is done by sunlight, heat, radiation, filtration and vibration. Chemical methods sterilization is done by chemicals like alcohol, aldehyde and organic chemicals. Coming to the topic sterilization by sunlight, it helps to sterilize most of the things in nature. The microbicidal activity of sunlight is due to the presence of the ultraviolet Rays which are present. So it is responsible for spontaneous sterilization in natural condition. For example, sterilization of, various water bodies which are there. Then it is more effective in killing germs due to the combination of ultraviolet Rays and the heat which is generated. Since it does not kill the Spores which are present, it is not a perfect sterilization method. Next is sterilization by heat. Now the use of heat is the most reliable method of sterilization. The killing effect is due to the denaturation and coagulation of protein. Now this method is used depends on the number of microorganisms present, characteristic of microorganism and the type of material. The methods of heat sterilization are again divided into two types. That is, Moist heat sterilization and dry heat sterilization. Moist heat sterilization involves the use of moisture to Sterilize various items dry heat sterilization is a method of Sterilization which basically works on the principle of conduction. Moist heat Sterilization can further be carried out at different temperatures that is below 100 degrees Celsius at 100 degrees Celsius and above 100 degrees Celsius. Sterilization by dry heat can be done by red heat, Flaming, incineration, and hot air oven. The moist heat sterilization at a temperature below 100 degrees Celsius. The method carried out over here is pasteurization. This method used for sterilization of milk and also to sterilize various serums and heat sensitive materials. Here sterilization is achieved by two methods so the first one is Holder method where the temperature is maintained at 63 degrees Celsius for 30 minutes. Next one is flash method where the temperature is maintained at 72 degrees Celsius for 15 to 20 seconds followed by quick cooling to 15 degrees Celsius or lower temperatures. Now this method destroy all the forms of sporulating pathogens, but it is not suitable for spore producing bacteria. Next is vaccine Bath in this process basically is used to decontaminate bacteria which are present in the vaccine by inactivating them. The vaccines are heated in the water bath at 60 degrees Celsius for one hour vegetative cells of the bacteria are killed but spores will survive. Next is Serum Bath. Here the contaminating bacteria in the serum preparation can be inactivated by heating in a water bath at 56 degrees Celsius for one hour on several successive days. Her proteins in the serum will coagulate at higher temperature. vegetative cells of the bacteria are killed, but spores will survive. Next is inspissation this technique is basically used to disinfect egg or serum containing media. The media is placed in the slopes of the Inspissator and they are heated at 80 to 85 degrees Celsius for 30 minutes on three successive days. On the first day, the vegetative bacteria would be killed. Those spores which germinate by the next day, are then killed the following day. Next is moist heat steriliization that is the

temperature at 100 degrees. Here boiling method is used. Boiling water at 100 degrees Celsius will be cleaning most of the vegetative bacteria and viruses. Some bacterial spores are resistant to boiling and they will survive, hence this is not a suitable method for sterilization. From here metal, Articles and glass wares can be disinfected by placing them inside the boiling water for 10 to 20 minutes and lid of the boiler should be close well during this period. Sterilization at temperature 100 degrees Celsius, so steam at atmospheric pressure is used here to sterilize substances i.e free stream and temperature at 100 degrees Celsius. is used to sterilize various instruments. so the steamer is used. The steamer is a metal cabinet with perforated trays to hold the articles and a conical lid to close. The bottom of the steamer is filled with water and heated steam sterilizes the articles when exposed to a period of 90 minutes, vegetative bacteria are killed in the first exposure and the spores that germinate by next day are killed on subsequent days. Moist heat sterilization that is temperature at 100 degree Celsius, steam under pressure. Over here Autoclaving Method is used now the principle which is followed over here is as a pressure increases the temperature increases i.e when a pressure inside a closed vessel increases the temperature at which water boils also increases, there by killing the microbes present at a temperature 121 degrees Celsius at 15 psi with 20 to 30 minutes holding time is commonly used. This is the image of the autoclave..Now basically, autoclaves are used to sterilize culture media, distilled water, and glasswares, item like Petri dishes, plastic tubings, cotton filters etc are wrapped in the brown paper or any other paper before putting in a autoclave. The mouth of the test tubes, flask, pipettes are to be plugged with a nonabsorbent cotton plug. Then all these are loaded in an autoclave with adequate space is maintained between the items for free circulation of the steam. Autoclave at 15 psi at 121 degrees Celsius for 20 to 30 minutes allow the pressure to drop gradually to zero psi and then only open the autoclave and unload tighten the caps of the bottles immediately transfer the glassware to the oven at 50 degrees Celsius to remove moisture and complete drying, then transferred to dust free sterilize Cabinet. Coming to the dry heat sterilization. The first method which is there is red heat now articles search as bacteriological loops, straight wires, searing spatulas are sterilized by holding them in a bunsen burner flame, till they become red hot. Then they are cooled and used further for inoculation. This is the image of the Bunsen Burner. Next is Flaming the articles to be sterilized. Just pass the articles a few times over the Bunsen Burner Flame and do not heat it to Red hot. We can Sterilize mouth of test tubes, flasks etc.Next is incineration. Now this is a method of destroying contaminated materials by directly burning them in an incinerator. This method results in the loss of articles. Next is hot air oven here articles are exposed to high temperature at 160 degrees Celsius for an hour in an electrically heated oven. Heat distribution is done with the help of a fan which is fitted inside at the bottom. articles sterilize include metallic instrument, glassware, swabs, oil, Grease and some pharmaceutical products. This is a image of the hot air oven. Next is sterilization by radiation. The radiation are basically of two types non ionizing and ionizing, so non ionizing Rays which are there are basically low energy rays with poor penetrating power example infra red Rays and UV rays. Ionizing Rays are high energy Rays with good penetrative power. Example are gamma Rays. Now coming to the non ionizing Rays that infra red Rays and UV rays. These are electromagnetic radiations with wavelengths longer than those of visible light. Infrared Rays bring about sterilization by generation of heat. It's articles to be sterilized are place in a moving conveyor belt and pass through a tunnel that is heated by infrared radiators to a temperature of 180 degrees celcius for 7.5 minutes used for rapid mass sterilization of

syringes, instrument and Glassware. Now UV rays are used for disinfecting enclosed areas such as laboratories, inoculation rooms and operation rooms. UV radiation is a form of UV tube or UV lamp is used in laminar air flow cabinet or inoculation chamber. working areas are exposed to UV radiation for 25 minutes before starting the work. This is the image of the laminar air flow. Disadvantages it has low penetration power, limited life of the UV bulb. Some bacteria have the DNA repair system. Some does not penetrate glass, paper or plastic. Eye protection is needed since it can cause severe damage. Next is the ionizing Rays i.e the gamma Rays. This method is also called cold sterilization, as there is no appreciable increase in temperature sterilization is accomplished in few seconds. Radiations with very high penetrating power. Highly lethal to DNA and other vital cell constituents. Cause biological damage by producing hyper reactive ions. Gamma Rays are used for sterilizing plastics, syringes, dressing packs, swabs, culture plates etc. Next is sterilization by filtration. filtration is basically used for the Heat sensitive substances like enzymes, antibiotics, amino acid, vitamins etc which cannot be sterilized by heating or at the high temperatures. contaminants like bacteria are filtered out by using millipore filters. Filters are made up of cellulose acetate or polycarbonates that contain small size pores nearly of 0.22 micron. The filters are placed in the filtration assembly and sterilized by autoclaving prior to use. The solution to be sterilized is passed through the Filtration Assembly and collected in a sterilized container. This is a image of the filters and the filtration assembly. Moving on to the types of filters. There are different types of filters which are available. Earthenware filters. They are made up of diatomaceous earth usually baked in the shape of a candle. Next is asbestos filters. They are made up Of chrysotile type of asbestos chemically composed of magnesium silicate. Pressed to form a disc and can be used only once. Disc is held inside a metal mount which is sterilized by autoclave. Next is sintered glass filters. They're basically made from the fine ground glass that is fused sufficiently to make small particles adhere to each other. Available in the form of disc fused into a glass funnel. Filters of grade five have average pore diameter of 1 to 1.5 microns. Washed in a running water in reverse direction and clean with warm, concentrated sulfuric acid. Sterilized by autoclaving. Next are the membrane. Filters. These are basically made from cellulose nitrate, cellulose acetate, polycarbonate and polyester. The pore diameter ranges from 0.015 Micron to 12 microns. The filters are sterilized by autoclaving. The advantages of the membrane filters are its known porosity, reusable after autoclaving and compatible with many chemicals. Now coming to sterilization by vibrations. Sonic and ultrasonic vibration are used to carry out sterilization. use high frequency sound waves of frequency greater than 20,000 cycles per second are generated using a sonicator which are basically used to disinfect various things. These vibrations are known to kill bacteria by causing cell disruption. They are used to clean and disinfect various equipments. It is not a reliable method. since many viruses are not killed. So in this module we have learned about the various physical methods of sterilization. These are the references to which I have referred to prepare this module. Thank you.