Sterilization

The terms **sterilization** and **disinfection** are used to indicate the treatment of material so as to destroy or otherwise eliminate any living or organisms present. However, sterilization is used where physical methods are used and disinfection is used where chemical agents are made use of.

Methods Commonly used for Sterilization

The methods used commonly in practice are:

1- *Killing organisms by heat:* Heat may be dry or moist.
2- *Destroying organisms by employing chemical antiseptics*, e.g. Lysol, phenol, perchloride of mercury, etc.
3- *Removing organisms mechanically by filtration:* e.g. Seitz, unglazed porcelain.

Sterilization by Heat

The time needed for sterilization is inversely related to the temperature of exposure – the higher the temperature, the shorter the time needed. High temperature kills bacteria by coagulating their proteins. Different types of bacteria show considerable differences in heat susceptibility. In general, vegetative forms are destroyed at lower temperature, whereas high temperatures are needed for sporing organisms.

Dry Heat

This is the preferred method for sterilizing glassware, e.g. of glass syringes and of materials such as oils, jellies and powders which are impervious to steam. Dry heat requires a much higher temperature or a much longer time at the same temperature than does moist heat. Dry heat can be used in the following ways:

Flaming: the articles are passed through the Bunsen flame, without letting them become red hot. It is used for scalpels, needles, mouths of culture tubes, glass slides, coverslips and points of forceps. Only the surfaces actually touched by the flame are sterilized.

Red Heat: Platinum loops, inoculating wires and needles are heated in the Bunsen flame until red hot.

Hot Air Oven: these are electrically heated and thermostatically controlled. Air circulates within the oven by convection currents. Suitable sterilizing times in the hot air oven are 3 hours at 140℃, 1 hour at 160℃ and 30 minutes at 180℃. All dry glassware, such as test tubes, petri-dishes, flasks, pipettes and throat swabs, etc. are made sterile by using hot air oven.

The method is not suitable for sterilizing culture media, liquids, rubber connections, glass to metal fitting and fabrics, e.g., masks, towels or gowns.

Moist Heat

***Temperature:** A temperature of 60-65℃ kills most vegetative bacteria (made use of in pasteurization of milk and preparation of vaccines).

****Boiling:** Boiling is frequently used for sterilizing syringes etc. but is not adequate as many spores withstand this temperature.

*****Steam:** Steam is the most effective technique of moist heat sterilization. Steam may be employed in three ways:

1- ***Steam at 100° C:*** The apparatus used commonly is called Koch's steamer. It has a vertical metal cylinder with a conical lid. It is fitted with a thermometer and has a small opening for escape of steam.

Sterilization by free steam can be done in two ways:

Prolonged Exposure: For $1^1/_2$ hours, used for broth or nutrient agar.

Intermittent Heat or Tyndallisation: It involves exposure for 20 minutes on three successive days and is used to sterilize sugars and gelatin which decompose on higher temperatures.

Principle: Spores would germinate after first steaming and destroyed on the next, three steaming would eliminate all spores and their vegetative forms.

2- ***Low Temperature Steam*:** This method is employed for sterilizing materials (blankets, polythene tubing, etc.) which would be damaged at higher temperatures.

3- ***Steam at Temperature above 100˚C (Autoclaving):*** The principle is that water boils when its vapors pressure is equal to the pressure of the surrounding atmosphere. If the pressure is raised inside a closed vessel, the temperature at which water boils will rise above 100˚C. At 15 Ibs. Pressure water boils at 120˚C. (If the autoclave pressure is taken down very quickly-the fluid-filled bottles may burst).

Timings:

10 Ibs. Pressure for 10 minutes – culture media.

15 Ibs. Pressure for 20 minutes – infected material.

20 Ibs. Pressure for 30 minutes – rubber gloves.

****Inspissation:** Used to sterilize serum containing media e.g., Loeffler's for diphtheria and Dorset's, or Lowenstein's media for TB. The inspissator consists of a double – walled copper box, with water flowing between 75 – 80˚C thermostatically. Sterilization is done for 2-3 hours on each of 3 successive days. A higher temperature may cause bubbling of the surface of the media.

Ultraviolet Radiation

Take care that the UV rays do not directly enter the eyes. Gram – negative bacteria are destroyed more rapidly than resistant and susceptibility of viruses is variable.

Ionizing Radiations

Cathode rays and gamma rays are the most effective and are being increasingly used to sterilize disposable items. These radiations have considerable disinfectant action.

Filtration

It may be used for the preparation from cultures of cell-free bacterial products, e.g. toxins and enzymes, to free virus containing fluids from bacteria and for the sterilization of media or media ingredients which would be damaged by heating. For this purpose, filters with pores sufficiently small to hold back bacteria must be employed. Filtration is usually carried out under negative pressure, the fluid being sucked through the filter into a receiving flask, which is connected to an exhaust pump. During filtration, the filter surface may adsorb material carrying an opposite charge – the material adsorbed may be the one desired in the filtrate.

Chemical Sterilization

Chemical agents can exert bactericidal or bacteriostatic effect. The bactericidal agents in lower concentrations exert bacteriostatic effect. The bactericidal effect is probably because of enzyme inactivation either by protein denaturation, oxidation or by a combination of the antibacterial agent with specific groups of enzyme proteins.

Various chemicals used are :

1- *Chloroform (volatile antiseptic):* used in preservation of serum for culture media at 0.25% concentration. Can be removed by heating to 56℃ .

2- *Phenol group:* Cresol, Lysol (strong antiseptics) are mainly employed for surgical instruments, discarded routine cultures, and pipettes, slides, etc., and disinfecting hands. Phenol 0.5% is used for preserving sera and vaccines.

3- *Metallic salts:* Perchloride of mercury in 1:1000 strength solution.

4- *Glycerol:* A 50% solution is used for preservation of certain viruses. Glycerol also kills contaminating organisms.

5- *Formalin:* It is the only method for sterilizing Perspex or polythene and for killing culture on plates. Can also be used for fumigation purposes.

6- *Sulfur:* Burning in air forms sulfur dioxide (SO_2) for fumigation.

7- *Halogens:* Chlorine and iodine. Chlorine for water disinfection and iodine for skin.

8- *Acids and alkalines:* Most bacteria grow in pH range of 5 to 9 and many grow at pH 7. Strong acids and alkalines can be used to disinfect contaminated materials.

9- *Alcohols:* These are used for disinfecting skin before injecting and before operations.

10- *Two groups of dyes (a) the aniline dyes and (b) the acridines* have been widely used as skin and wound disinfectants.

11- *Quaternary ammonium compounds* are active against both gram-positive and gram-negative species but are not effective against spores and *Mycobacteria.* They are used for sterilizing food utensils in restaurants and hotels and for disinfecting blankets in hospitals.

12- *Glutaraldehyde (Cidex)* affects even spores and *Mycobacteria*. It is employed as 2% solution and is recommended for sterilizing cystoscopies, etc.

13- *Ethylene oxide gas* is being widely used to sterilize disposable plastic syringes, petri dishes, etc.

Antiseptics

The word " antiseptic" has acquired the special meaning of an antimicrobial agent (microbicidal / microbistatic), suitable for application to living tissues and intended to reduce the viable count or inhibit the growth of microbial flora. Skin antisepsis has moved on form the confines of hospital care and treatment to now play an essential role in secondary and tertiary healthcare setups, thereby also going a large footage in home care and maintenance of general personal hygiene.

****Q/ Explain both of general and specialized antisepsis with examples?**