STERLISATION METHODS
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WHAT IS STERILIZATION?

- Sterilization can be defined as a process by which all viable forms of micro-organisms(such as fungi, bacteria, viruses) are removed or destroyed.
- Micro organisms are present at everywhere. Since they cause contamination, infection, and decay, it becomes necessary to remove or destroy them from materials or from areas.
- Sterilization procedures used is should be simple but effective and of relatively short duration.

Physical methods

1) **Sunlight**

2) **Drying**

3) **Heat:-** i) Moist heat

ii) Dry heat

4) **Radiation**

1) Sun light - It is responsible for spontaneous sterilization in natural conditions.

- In tropical countries, the sunlight is more effective in killing germs due to combination of ultraviolet rays and heat.
- By killing bacteria suspended in water, sunlight provides natural method of disinfection of tanks and lakes.

2) Drying - Moisture is essential for growth of bacteria.

- Drying in air has dangerous effect on many bacteria.
- However, spores are unaffected.
- Therefore, it is not satisfactory method for sterilization.

3) Heat sterilization- it has also **two** type

A) **Dry heat sterilization-** In dry heat sterilization, dry heat is used for sterilizing different materials. Heated air or fire is used in this process. As compared to the moist heat sterilization, the temperature is higher. The temp. is maintained for almost an hour to kill the most difficult of the resistant spores.

Dry heat sterilization also have **4** type:- 1) Hot air oven

2) Red hot sterilization

3) Flaming

4) incineration

1) **Hot air oven:- Hot air ovens** are electrical devices which use dry heat to **sterilize**. They were originally developed by pasteur.

- Generally, they can be operated from **50 to 300 °C**, using a thermostat to control the temperature. ... An air circulating fan helps in uniform distribution of the heat.

2) **Red hot sterilization:-** Sterilization by holding them in Bunsen flame till they become red hot. It use for bacteriological loops, straight wires, tips of forceps & spatulas

3) Flaming:- This is a method of passing article over a flame, but not heating it to redness.

Use- scalpels, mouth of test tubes, flask, glass slide & coverslips

4) Incineration:- Incineration is a waste treatment process that involves of organic substances contained in **waste materials.**

- This method also burns any organism to ash.

It is **used** to **sterilize** medical and other biohazardous waste before it is discarded with non-hazardous waste

sensitive materials and materials through which steam is permeable. culture media is also **sterilized** through **moist heat sterilization.**

It has also 3 types:- 1) Below 100°C 2) Above 100°C 2) At 100°C

1) **Below 100°C**:- It has also 2 types

i) Pasteurization- Its purpose is to reduce the bacterial population of a liquid such as a milk & destroy Organism that ma cause spoilage.

Spores are not affected by this method.

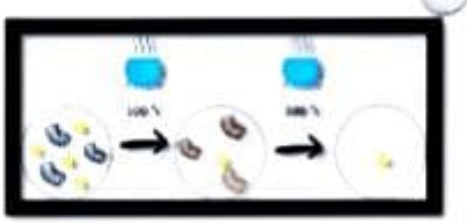
ii) inspissation:- Heating at 80-85°C for half an hour daily on three consecutive days.

Serum or egg media are sterilized.

2) **At 100°c**:- It has also 2 types.

i) Boiling:-**Boiling** is a very simple method of water **disinfection**. Heating water to a high temperature, 100°C, kills most of the pathogenic organisms, particularly viruses and bacteria causing waterborne diseases. In order for **boiling** to be most effective, the water must **boil** for at least 20 minutes.

ii) Tyndallization:- Exposure at 100°C for 20-45 minutes for 3 successive days. Used for sterilizing sugars, gelatin, & serum containing media.



3) **Above 100°C**:- Autoclave

- Autoclaving is the most reliable method for sterilization.
- Autoclave use pressurized steam to destroy microorganism, and are the most dependable system available for the decontamination of laboratory waste & the sterilization of glass wares, media, & reagents.

- For efficient heat transfer, steam must flush the air out of the autoclave chamber.
- Generally the conditions employed are temperature up to 121-134° for 15-20 minutes under 15 lbs pressure. The condition based on the type of the material used for sterilization.

4) Radiation sterilization:- It has 2 types.

A) Non-ionizing(Hot sterilization):-

i) Infrared rays- used for rapid mass sterilization of prepacked items such as syringe, catheters(a thin tube that is put into the body in order to remove liquids)

ii) U.v. rays- used for disinfecting enclosed area such as entry ways, operation theatres & labs.

B) Ionizing(Cold sterilization):-

i) Gamma rays & X-rays:- Used for sterilizing plastics, syringes, swabs, animal feeds, oils, greases, fabric & metal foils

- **Advantages of radiation sterilization**:- clean & dry process, ensure full exposure of object from all direction.
- **Disadvantages**:- posses threat to human, lengthy process, requires very qualified person

Chemical methods

1) Gaseous sterilization:- i) Ethylene oxide

ii) Formaldehyde gas

2) Liquid sterilization:- i) Alcohol

ii) Phenol

I. **Ethylene oxide**:- EtO sterilization is mainly use to sterilize medical & pharmaceutical products that can not support conventional high temperature steam sterilization- such as devices that incorporate electronic components, plastic packaging or plastic container.

- This method uses automatic device filled with ethylene oxide gas at temperature below 100°C to sterilize complex & delicate material.
- EtO destroys microorganism by chemically reacting with nucleic acid.

- Advantages :- Fully automatic, high efficiency, 100% result
- Disadvantages:- Complex and time consuming process, carcinogenic safety concern

II. Formaldehyde gas:- Another low temperature method for sterilizing heat sensitive items is formaldehyde sterilization. Formaldehyde is an organic chemical compound which is a by-product of the metabolism of many organisms and is commonly found in fresh air, rainwater, foods, industrial products and fabrics.

- It is considered even more dangerous than EtO and is therefore less commonly used for sterilization.

- Formaldehyde sterilization is used where sterilization by steam or high temperature is not possible
- Formaldehyde is soluble in water and its inactivation power is greatly improved by the presence of humidity. It is most commonly used as a disinfectant, but sometimes formaldehyde is used as a sterilizing agent. The process is known as **low temperature steam and formaldehyde** (LTSF)
- In countries such as united kingdom, germany, sweden, denmark and norway sterilization by LSTF is accepted, but not common. On the other hand in several countries formaldehyde as a sterilizing agent is discouraged. LTSF has not been FDA cleared for use in healthcare facilities in the USA.

- Advantages:- Very reactive molecule, Faster cycle time compared to EtO, cost per cycle is lower than EtO, after sterilization most loads are available for immediate use
- Disadvantages:- The vapour is extremely irritating to the eyes, weak penetrating power compared to EtO, operates on a higher temperature than EtO, formaldehyde residue can remain on the sterilized goods if the rinsing phase is not 100% efficient. This can be harmful for the patients

2) Liquid sterilization

I) Alcohol:- Alcohols are effective disinfectants for many reasons. They evaporate quickly, without leaving a residue. They are capable of dissolving lipids, which makes them effective against lipid-wrapped viral cells such as HIV and hepatitis A. They are inexpensive and relatively easy to handle, although their vapors are flammable.

- Ethanol and isopropyl alcohol are both members of the alcohol family and have similar disinfectant properties. Ethanol is the type of alcohol present in alcoholic beverages. Isopropyl alcohol is also known as isopropanol, 2-propanol or rubbing alcohol. When used as disinfectants, both are typically at a concentration of 70 percent in water.

- phenols acts by damaging cell membrane thus releasing cell contents & causing lysis.
- phenols is commonly found in mouth washes, scrub soaps, & surface disinfectants.
- phenols are used for decontamination of the hospital environment, including laboratory surfaces, & non critical medical items.
- Examples:-

Filtration (Mechanical) methods

- Filtration sterilization used for heat sensitive materials to sterilize.
- Filtration process does not destroy but removes the microorganisms.
- Filtration allows for the exclusion of organisms based upon size.

❑ **Procedure:-** The solution to be sterilized is passed through the filter and collected in the sterile receiver by the application of positive pressure to the nonsterile compartment or negative pressure to the sterile slide.

Mode of action

The filters are thought to function by one or usually a combination of the following:

1. Sieving or screening
2. Entrapment
3. Electrostatic attraction

➢ When a particle is larger than the pore size of the filter the particle is retained on the filter- this known as **sieving or screening**

- **Entrapment** occurs when a particle smaller than the size of the pore enters into the pore channel and lodges onto the curves of the channel while passing through it.
- **In Electrostatic attraction** Particles are attracted & absorbed at the surface of the filter bed which is oppositely charged.
- There are 4 types of filters:-

1. Membrane filters
2. Sintered or Fritted glass filters
3. Seitz filters
4. Ceramic filters

or nitrate). They are very fine. They are fixed in some suitable holders.

- Nominal pore size is 0.22 ± 0.02mm or less is required.
- The membranes are brittle when dry. In this condition they can be stored for years together. They become very tough when dipped in water.
- They are suitable for sterilizing aqueous and oily solutions but not for organic solvents such as alcohol, chloroform etc.

- They are sterilized by autoclaving or by ethylene oxide gas. They can not be sterilized by dry heat as they decompose above 130°C.
- Membrane filters are generally blocked by dirt particles and organisms. Pre-filtration (through glass-fibre paper prefilter) reduces the risks of membrane filter.
- Examples:- i) MF-Milipore - it is a mixture of cellulose esters
  ii) Sartorius regular – it is made of cellulose nitrate