

STERILIZATION AND DISINFECTION

By

Dr. Ali Aldeewan

INTRODUCTION

- ▣ Microorganisms are the agents of contamination, infection, and decay.
- ▣ Hence it becomes necessary to remove them from materials and areas.
- ▣ Early civilization practiced salting, smoking, pickling and exposure to sunlight .

In mid 1800s Lister developed Aseptic techniques to prevent contamination of surgical wounds. Prior to this development:

- Nosocomial infections caused death in 10% of surgeries.
- Up to 25% mothers delivering in hospitals died due to infection.

DEFINITION

STERILIZATION

The process of freeing an article from microorganisms including their spores.

DISINFECTION:

Reducing the number of pathogenic microorganisms to the point where they no longer cause diseases.

Sepsis: Comes from Greek for decay or putrid.
Indicates bacterial contamination.

Asepsis: Absence of significant contamination.

- ▣ **Aseptic techniques** are used to prevent contamination of surgical instruments, medical personnel, and the patient during surgery.
- ▣ **Aseptic techniques** are also used to prevent bacterial contamination in food industry.

Bacteriostatic Agent:

An agent that *inhibits* the growth of bacteria, but does not necessarily kill them.

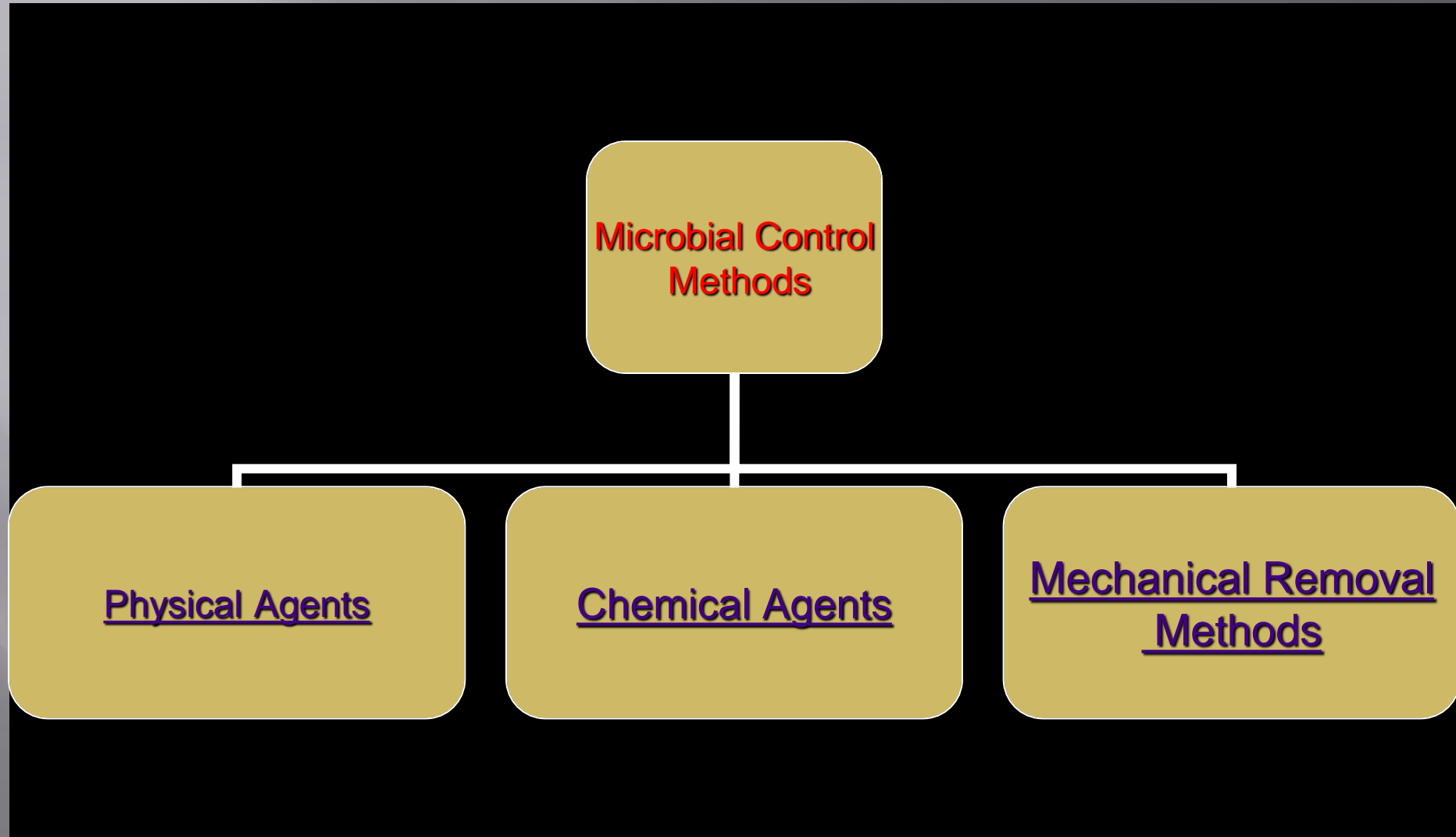
◆ **Bactericide:**

An agent that kills bacteria. Most do not kill Endospores.

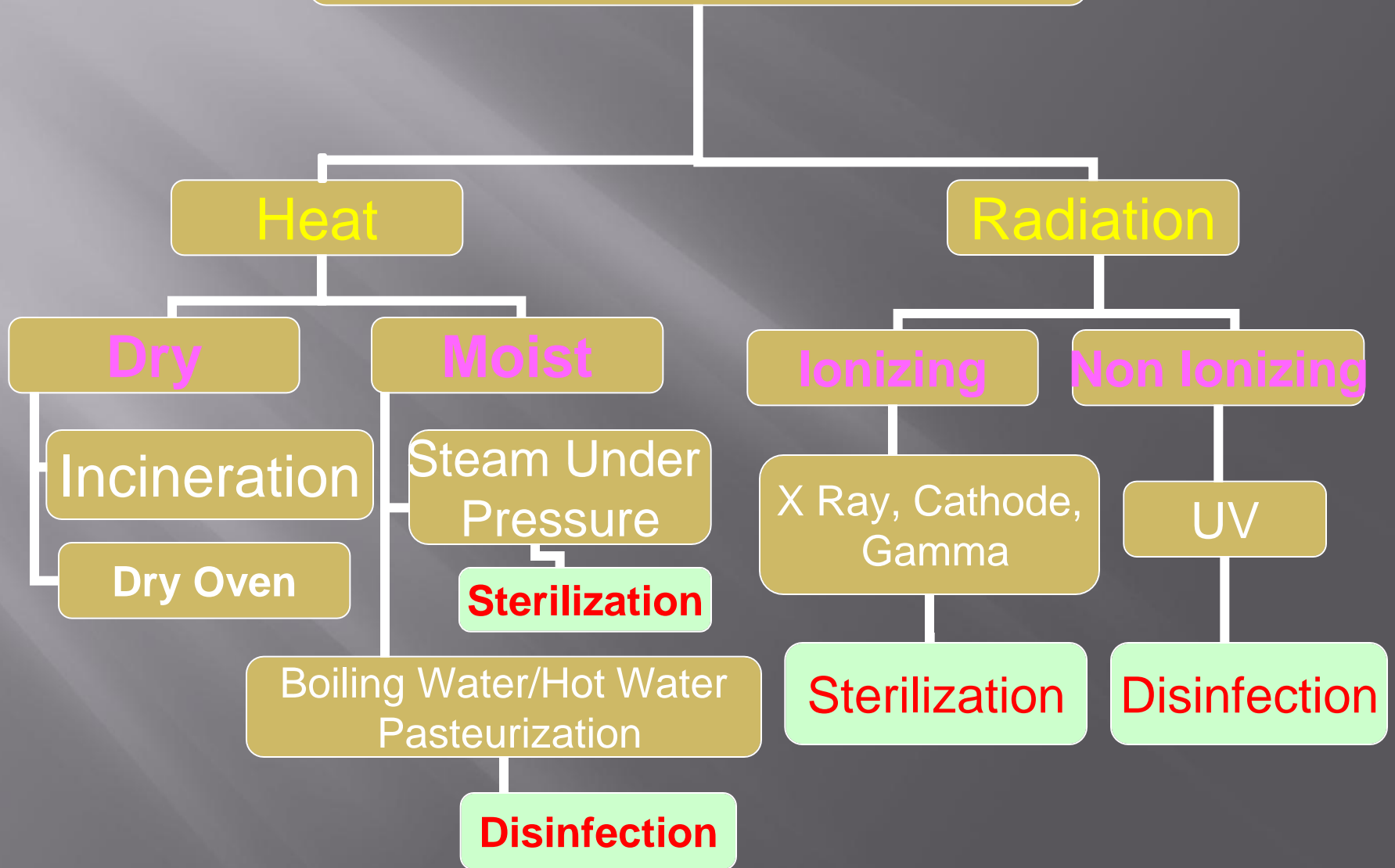
◆ **Sporicide:**

An agent that kills spores.

Methods of Sterilisation



Physical Agents



Chemical Agent

Gas

Liquids

Sterilization

Disinfection

Animate

Inanimate

Chemotherapy

Antiseptics

Sterilization

Disinfection

Mechanical Removal Methods

```
graph TD; A[Mechanical Removal Methods] --> B[Filtration]; B --> C[Air]; B --> D[Liquids]; C --> E[Disinfection]; D --> F[Sterilization];
```

Filtration

Air

Liquids

Disinfection

Sterilization

Physical Methods of Sterilisation

Sterilisation By Dry Heat:



Hot Air Oven

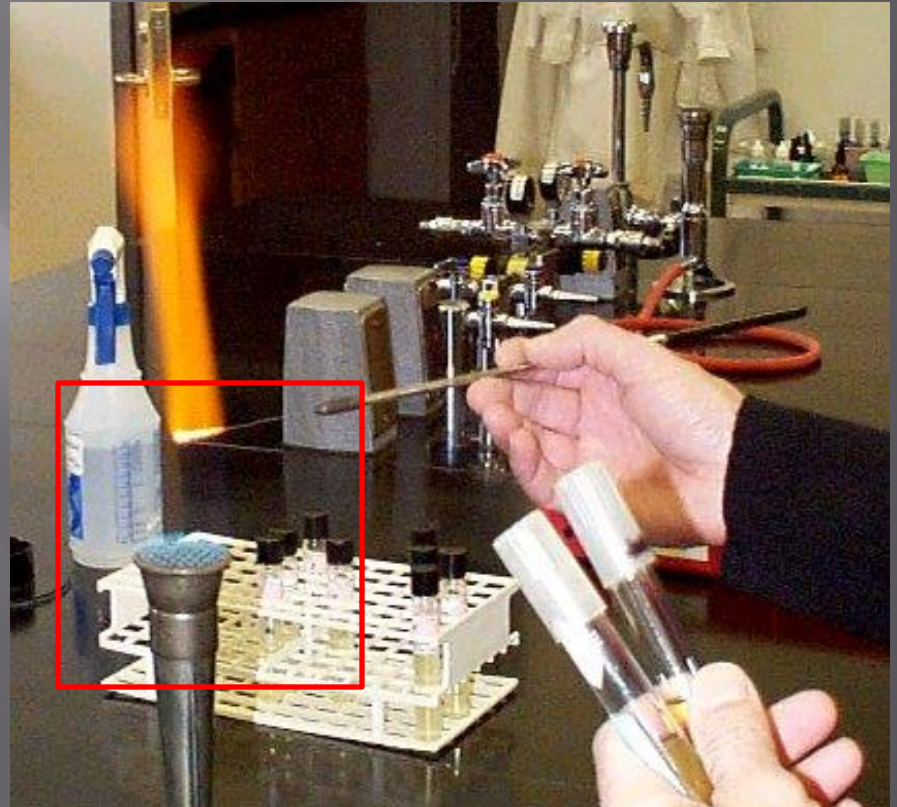
- Kills by oxidation effects
- The oven utilizes dry heat to sterilize articles
- Operated between 50°C to 250/300°C.
- A holding period of 160°C for 1 hr is desirable.
- There is a thermostat controlling the temperature.
- Double walled insulation keeps the heat in and conserves energy,

Uses:

- ▣ To sterilize Forceps, Scissors, Scalpels, Swabs.
- ▣ Pharmaceuticals products like Liquid paraffin, dusting powder, fats and grease.

FLAMING

- ▣ Inoculation loop or Wire, the tip of Forceps and spatulas are held in a bunsen flame till they are red hot.



Incineration

This is an excellent method of destroying materials such as contaminated cloth, animal carcasses and pathological materials.

MOIST HEAT STERILIZATION

Kills microorganisms by **coagulating** their proteins.

Moist heat sterilization is carried out with following methods

- ▣ Temp below 100°C: “Pasteurization”, Inspissator.
- ▣ Temperature at 100°C: Boiling.
- ▣ Steam at atmospheric pressure: Koch/Arnold's steamer.
- ▣ Steam under pressure: Autoclave.

Pasteurisation

- ▣ Process of killing of pathogens in the milk but does not sterilize it .
- ▣ Milk is heated at 63°C for 30 mins.
(Holder method)
- ▣ At 72°C for 15-20 Sec. Rapid cooling to 13°C
(Flash process)



Water Bath

HOT WATER BATH

- ▣ To inactivate non sporing bacteria for the preparation of vaccines - **Special vaccine bath** at 60°C for one hour is used
- ▣ Serum or body fluids containing coagulable proteins can be sterilized by heating for 1 hr at 56°C in a water bath for several successive days.



Inspissator

INSPISSATOR

- ▣ Sterilizes by heating at 80-85°C for half an hour for 3 successive days
- ▣ Used to sterilize media such as Lowenstein-Jensen & Loeffler's serum

TEMPERATURE AT 100°C

Boiling:

Kills vegetative forms of bacterial pathogens.

- ▣ **Hepatitis virus:** Can survive up to 30 minutes of boiling.
- ◆ **Endospores:** Can survive up to 20 hours or more of boiling



Steam Sterilizer

STEAM AT ATMOSPHERIC PRESSURE

- ❑ Steam is generated using a steamer (Koch/ Arnold)
- ❑ Consists of a Tin cabinet
- ❑ Has a conical lid to enable the drainage of condensed steam
- ❑ Perforated tray above ensures materials are surrounded by steam.
- ❑ For routine sterilization exposure of 90 mints is used

- ▣ For media containing sugar and gelatin exposure of 100°C for 20 min for 3 successive days is used
- ▣ The process is termed as
Tyndallisation / Intermittent Sterilization

STEAM UNDER PRESSURE - AUTOCLAVE

- ▣ Works on the principle of **Steam under pressure**
- ▣ Invented by Charles Chamberland in 1879.



AUTOCLAVE

- ❑ **Autoclave** consists of a vertical or a horizontal cylinder.
- ❑ One end has an opening which is meant for keeping materials to be sterilized.
- ❑ The lid is provided with a Pressure gauge, to measure the pressure
- ❑ A safety valve is present to permit the escape of steam from the chamber
- ❑ Articles to be sterilized are placed in the basket provided
- ❑ Sterilisation is carried out under pressure at 121° for 15 mints.

CHEMICAL AGENTS

Chemical agents act by

- ▣ Protein coagulation
- ▣ Disruption of the cell membrane
- ▣ Removal of Sulphydryl groups
- ▣ Substrate competition

ALCOHOLS

- ▣ Ethanol / Isopropyl alcohol are frequently used
- ▣ No action on spores
- ▣ Concentration recommended 60-90% in water

Uses

- ▣ Disinfection of clinical thermometer.
- ▣ Disinfection of the skin - Venupuncture

ALDEHYDES

- Formaldehyde & Glutaraldehyde are frequently used
- Formaldehyde is bactericidal, sporicidal & has a lethal effect on viruses.
- Glutaraldehyde is effective against Tubercle bacilli, fungi and viruses

USES

FORMALDEHYDE

- ❑ To preserve anatomical specimens
- ❑ Destroying Anthrax spores in hair and wool
- ❑ 10% Formalin+0.5% Sodium tetra borate is used to sterilize metal instruments

USES

GLUTARALDEHYDE

- ▣ Used to treat corrugated rubber anesthetic tubes, Face masks, Plastic endotracheal tubes, Metal instruments and polythene tubing

DYES

Two groups of dyes are used

Aniline dyes

Acridine dyes

ANILINE DYES

- ▣ Are Brilliant green, Malachite green & Crystal violet
- ▣ Active against Gram positive bacteria
- ▣ No activity against tubercle bacilli

ACRIDINE DYES

- ▣ **Acridine dyes in use are orange in colour**
- ▣ **Effective against Gram positive than Gram negative**
- ▣ **Important dyes are Proflavine, Acriflavine, Euflavine**

HALOGENS

- ▣ **Iodine** in aqueous and alcoholic solution has been used widely as a skin disinfectant
- ▣ Actively bactericidal with moderate against spores
- ▣ **Chlorine** and its compounds have been used as disinfectants in water supplies & swimming pools

PHENOLS

- ▣ Obtained by distillation of coal tar
- ▣ Phenols are powerful microbicidal substances
- ▣ Phenolic derivatives have been widely used as disinfectants for various purposes in hospitals
- ▣ Eg: Lysol, cresol

USES

- ▣ Various combinations are used in the control of pyogenic cocci in surgical & neonatal units in hospitals.
- ▣ Aqueous solutions are used in treatment of wounds

GASES

Ethylene Oxide

- Colorless ,Highly penetrating gas with a sweet ethereal smell.
- Effective against all types of microorganisms including viruses and spores

USES

- ▣ Specially used for sterilizing heart-lung machines, respirators, sutures, dental equipments, books and clothing.
- ▣ Also used to sterilize Glass, metal and paper surfaces ,plastics, oil, some foods and tobacco.

FORMALDEHYDE GAS

- ▣ Widely employed for fumigation of operation theatres and other rooms

BETA PROPIOLACTONE

- ▣ Used in fumigation
- ▣ For sterilisation 0.2% BPL is used
- ▣ Has a rapid biocidal activity
- ▣ Very effective against viruses

SURFACE ACTIVE AGENTS AND METALLIC SALTS

Substances which reduce the surface tension –

Surface active agents

- ▣ Cations are widely used in the form of quaternary ammonium compounds.
- ▣ Markedly bactericidal, active against Gram positive organisms.
- ▣ No action on spores, tubercle bacilli, viruses

METALLIC SALTS

- ▣ **The salts of silver, copper and mercury are used as disinfectants.**
- ▣ **Act by coagulating proteins**
- ▣ **Marked bacteriostatic, weak bactericidal and limited fungicidal activity**

TESTING OF DISINFECTANTS

- ▣ Rideal Walker test
- ▣ Chick Martin Test

Sterilisation by filtration

Filtration helps to remove bacteria from heat labile liquids such as sera and solutions of sugar, Antibiotics.

The following filters are used

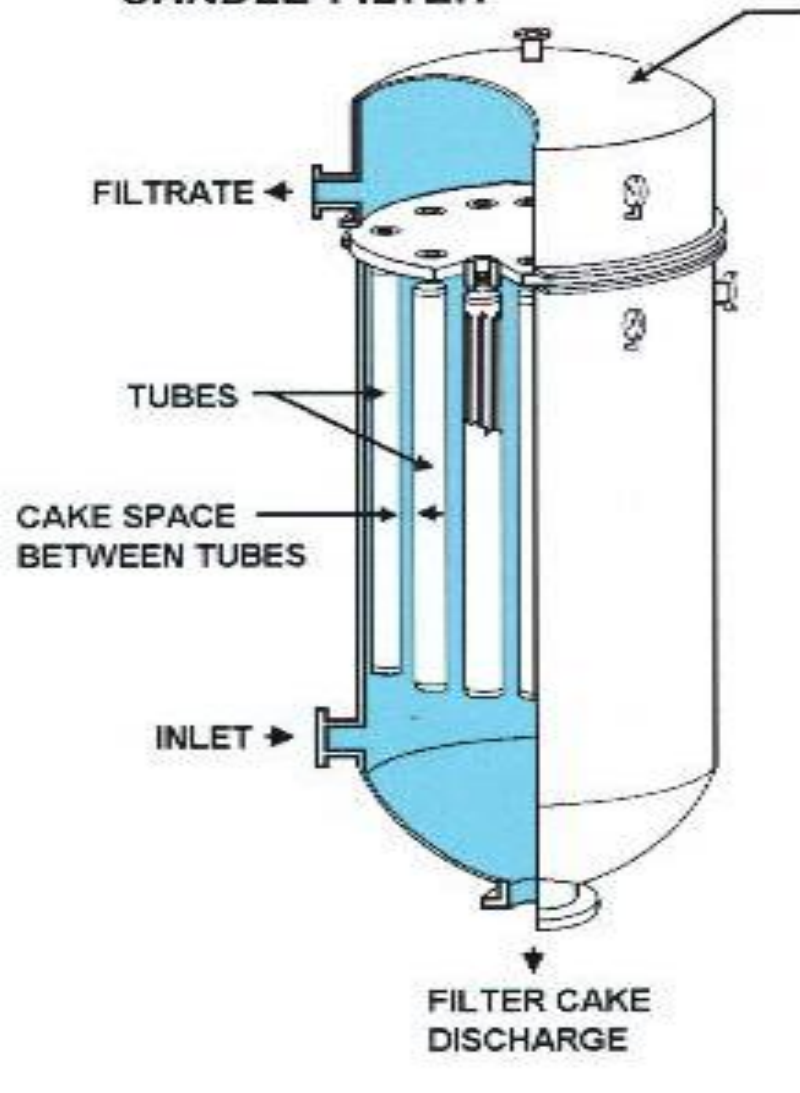
Candle filters

Asbestos filters

Sintered glass filter

Membrane filters

CANDLE FILTER



CANDLE FILTER

CANDLE FILTERS

- ▣ Widely used for purification of water

Two types

(a) Unglazed ceramic filter – Chamberland filter

(b) Diatomaceous earth filters – Berkefeld filter



SEITZ FILTER



ASBESTOS DISCS

ASBESTOS FILTER

- ▣ Disposable single use discs
- ▣ High adsorbing tendency
- ▣ Carcinogenic

Eg: Seitz filter



SINTERED GLASS FILTER

SINTERED GLASS FILTER

- ▣ Prepared by heat fusing powdered glass particles of graded size
- ▣ Cleaned easily, brittle, expensive.



MEMBRANE FILTER

MEMBRANE FILTERS

- ▣ Made of cellulose esters or other polymers

Uses

- ▣ Water purification & analysis
- ▣ Sterilization & sterility testing
- ▣ Preparation of solutions for parenteral use

RADIATION

Two types of radiations are used

NON -IONISING

IONISING

Non- Ionising radiation:

- ▣ Electromagnetic rays with longer wavelength
- ▣ Absorbed as heat
- ▣ Can be considered as hot air sterilization
- ▣ Used in rapid mass sterilizations of pre packed Syringes and catheters

Eg: UV rays

IONISING RADIATIONS

- ▣ X- rays, gamma rays & cosmic rays.
- ▣ High penetrative power
- ▣ No appreciable increase in the temperature –
COLD STERILISATION
- ▣ Sterilize plastics Syringes, catheters, grease fabrics metal foils

ULTRASONIC AND SONIC VIBRATION

- ▣ Bactericidal
- ▣ Microorganisms vary in their sensitivity, hence no practical value in sterilization and disinfection

THANK YOU