SNS COLLEGE OF ALLIED HEALTH SCIENCES

SNS Kalvi Nagar, Coimbatore - 35

Affiliated to Dr MGR Medical University, Chennai

DEPARTMENT OF OPERATION THEATRE AND ANAESTHESIA TECHNOLOGY - II YEAR

COURSE NAME: STERILISATION PROCEDURES

TOPIC – STERILISATION AND DISINFECTION

MRS.GAYATHIRI.K
Lecturer
OTAT
SNSCAHS

INTRODUCTION

BACTERIOSTATIC : agents only precent the multiplication of bacteria which may ,however

CLEANING : plays an important role in preparatory role by sterilization or disinfection by removing soil & other dirt and reducing the microbial burden making sterilization more effective.

DECONTAMINATION : Refers to the process of reducing an article or free of danger from contaminations including microbial, chemical ,radioactive and other hazards .

INTRODUCTION

STERILISATION : It is defined as the process of killing all microorganisms including bacteria , bacterial spores , fungi , and virus

DISINFECTION : it means the destruction or removal of all pathogenic organism capable of giving rise to infection

ANTISEPTICS : are disinfection that can be used on body surfaces such as the skin or vaginal tract to reduce the number of normal floral and pathogenic contaminants

BACTERICIDAL OR GERMICIDAL : agents can kill bacteria

METHODS OF STERILIZATION

PHYSICAL

Heat

DRY HEAT

- Red heat)
- Flaming
- Incineration
- Hot air oven

MOIST HEAT

- Below 100˚C (pasteurization)
- At 100˚C (Boiling)
- Above 100˚C (Autoclave)

Filtration

- Candle filter
- Asbestos filter
- Sintered glass filter
- Membrane filter

Radiation

- Ionizing (Gamma rays)
- Non Ionizing (UV rays)

CHEMICAL

- Alcohol
- Aldehydes
- Biguanides
- Beta
- Propiolactone
- Surfactants
- Phenol
- Halogens
- Metallic salt
- Peroxide
- Dyes
- Quaternary Ammonium compound

CHEMICAL METHOD :

- Surfactants
- Peroxides
- Alcohols
- Aldehydes
- Biguanides
- Dyes
- Betapropiolactons
- Halogens
- Metalic salt
- Phenolic compounds
- Quaternary ammonium compounds

CHEMICAL METHOD :

I. Disinfection
II. Gaseous sterilization

PROPERTICS OF AN IDEAL DISINFECTION :

1) Should have a wide spectum of activity and be effective against all microorganism
2) Should not be toxic to human tissue of
3) *Should be active* in presence of organic matter such as body fluid , blood pus
4) Should be compatible with other antiseptics and disinfectants.
5) Should be effective at all ph ranges .

CHEMICAL METHOD :

I. Disinfection
II. Gaseous sterilization

PROPERTICS OF AN IDEAL DISINFECTION :

1) Should have a wide spectum of activity and be effective against all microorganism
2) Should not be toxic to human tissue of
3) *Should be active* in presence of organic matter such as body fluid , blood pus
4) Should be compatible with other antiseptics and disinfectants.
5) Should be effective at all ph ranges .

FACTORS WHICH DETERMINE THE POTENCY OF A DISINFECTION :

Concentration of the disinfection	If the disinfection is diluted potency will be reduced
Time of action	disinfection process
Nature of organism :	Sporing bacteria may not be affected by
. PH of medium :	Some disinfection might get inactivated
Temperature	Increase in temperature increase the disinfection rate of reaction

CHEMICAL METHOD :

I. Disinfection
II. Gaseous sterilization

PROPERTICS OF AN IDEAL DISINFECTION :

- Should have a wide spectum of activity and be effective against all microorganism
- Should not be toxic to human tissue of
- Should be active in presence of organic matter such as body fluid , blood pus
- Should be compatible with other antiseptics and disinfectants.
- Should be effective at all ph ranges .

MECHANISM OF ACTION :

1. Coagulation of protein .

2. Lysis of cell membrane .

3. Substrate competition and interference with enzyme functioning .

4. Interaction with various functional groups .

CHEMICAL AGENTS	MECHANISM	USES
1. Alcohol (Ethanol , Isopropanol)	Denaturation of bacterial proteins	Skin antiseptics, surface decontamination of incubators & cabinet interiors, disinfection clinical thermometer .
2. Aldehydes (Glutaraldehyde , Formaldehyde)	Inactivation of bacterial proteins .	Preservation of biological specimens , destroying another spores in fumigation. Cold sterilization and fixation, surface decontamination, disinfection of hospital instruments requirements glass wares.
3. Biguanides (Chlorhexidine)	Damage plasma membrane	Skin & mucous membrane Disinfection .

4. Beta Propiolactone	**Damage DNA , RNA & cause Alkylation .**	**Fumigation , sterilization of biological products .**
5. Surfactants (Soap and detergents Na lauryl sulphate , benzalkonium chloride cetrimide .	Disruptor of cell membrane	Detergents and wetting agents
6. Phenol compounds (Phenol , Cresol, Lysol)	Damage to cell membrane , inactivation of proteins , oxidizes & dehydrogenases .	Disinfection in hospital
7. Halogens (Chlorine eg : Na hypochlorite, chloramines, hypochlorous, acid, iodine) Eg : Tincture iodine, Povidone iodine .	Oxidizing agent , Protein Denaturation	Surface decontamination emergency spill clean up disinfectant . Antiseptic surface decontanimation, Instruments disinfection

7. Peroxide (Hydrogen peroxide)	**Oxidizing agent**	Disinfection
8. Metallic salt (Silver, eg: silver nitrate , mercury) Eg : Methiolate , mercurochrome .	Combine with sulphydryl groups, coagulation proteins & inactivate enzymes .	Antiseptic to purent against gonorrheal infection in infants, Antiseptic on wound .
10. Dyes (Aniline dyes , malachite green acridine dyes, acriflavine, proflavine.	React with acid group in cell . Impair DNA& destroy reproductive capacity .	Selective agents in culture media, eg : LJ media skin antiseptic .
11.Quaternary Ammonium compounds (Zephiran , Triclosan)	Sulfate active agents (cationic detergent)	Surface decontamination & disinfection equipments .

TESTING OF DISINFECTION :

1. Testing of disinfectants : It compares the efficiency of a disinfectant with phenol .

PRINCIPLE :

- Equal quantity of organisms are subjected to the action of varying concentration of phenol & the disinfection to be tested .

- That concentration of disinfection needed to sterilize the organism in a given time divided by the corresponding concentration of phenol is called the phenol coefficient of that disinfectant .

- Any disinfectant with a phenol coefficient of 1 is equal to phenol as a disinfection . A phenol coefficient of greater than 1 means that the test disinfectant is better than phenol .

a) Deal walker test : The disinfectant dilution are made using distilled water & tested .

b) Chick martin test : The disinfectant are tested in presence of organic matter (dried yeast) .

2. Capacity dilution test : It involved adding of bacterial in oculars to the disinfected in 3 successive lots at 0,1 and 5minutes , this is the principle of capacity test where the capacity or lack of capacity of the disinfection to destroy the successive addition of bacterial culture is tested .

IN USE TEST :-

It is done for hospital disinfectants .

GASEOUS STERILANTS :-

ETHYLENE OXIDE :

- Highly explosive, expensive .
- Acts by denaturation of proteins and damage to DNA .
- Acts best at 50 – 55 ˚C and has a boiling point of 10.7 ˚C .
- It ia an excellent sterilizing agent for those materials destroyed by heat .
- Its disadvantages: carcinogen & mutagen
- used for the sterilization of
  - heart lung machine
  - Respirators
  - Sutures
  - Dental equipment
  - Clothing

FORMALDEHYDE GAS :-

- Acts against amino group of protein molecules & inactivates them.
- It has a pungent odour & is irritated to the skin , eyes and respiratory tract .
- 37% aqueous formaldehyde (formalin) is used to preserve biological specimens & inactive viruses & bacteria in vaccines .

PHYSICAL AGENTS

HEAT :-

- Most reliable method of sterilization factors affecting heat as a method of sterilization .
- Types of heat : - Dry heat
- - Moist heat
- Temperature and time which are inversely proportional .
- Type of article to e sterilized
- Numbers & type of organism .

DRY HEAT :-

- Acts by oxidation of essential components of the cell

FLAMMING :-

PRINCIPLE : Passing the object through the flame of Bunsen burner without heating to redness used for sterilization of

- Glass slides
- Mouth of culture tube .

INCINERATION :-

Temperature : > 1000 ˚C

Principle : Infection materials is covered to sterile ash by burning in incineration .

Used for : Destruction of contaminated disposable materials (waste) .

HOT AIR OVEN

- Most widely used method of sterilization using dry heat.
- **PRINCIPLE :** The killing effect is brought by denaturation of enzymes & proteins and oxidation of essential all components.
- **Structure & process :** It comprises of an electrically heated , insulated chamber fitted with insulated door.
- The chamber is usually of stainless steal & the inner surfaces are polished to minimize heat loss.

HOT AIR OVEN

- The oven is heated electrically with heating elements in the wall of chambers.
- Heat id delivered to the load primarily by connection & radiation.
- A fan is fitted for efficient air circulation & heat distribution within the oven.
- It is also fitted with thermostat to control the temperature.
- The temperature various between 160˚C – 170˚C & the duration of 1-2 hrs.
- Ample spacing must be allowed in the oven to permit free circulation of hot air .

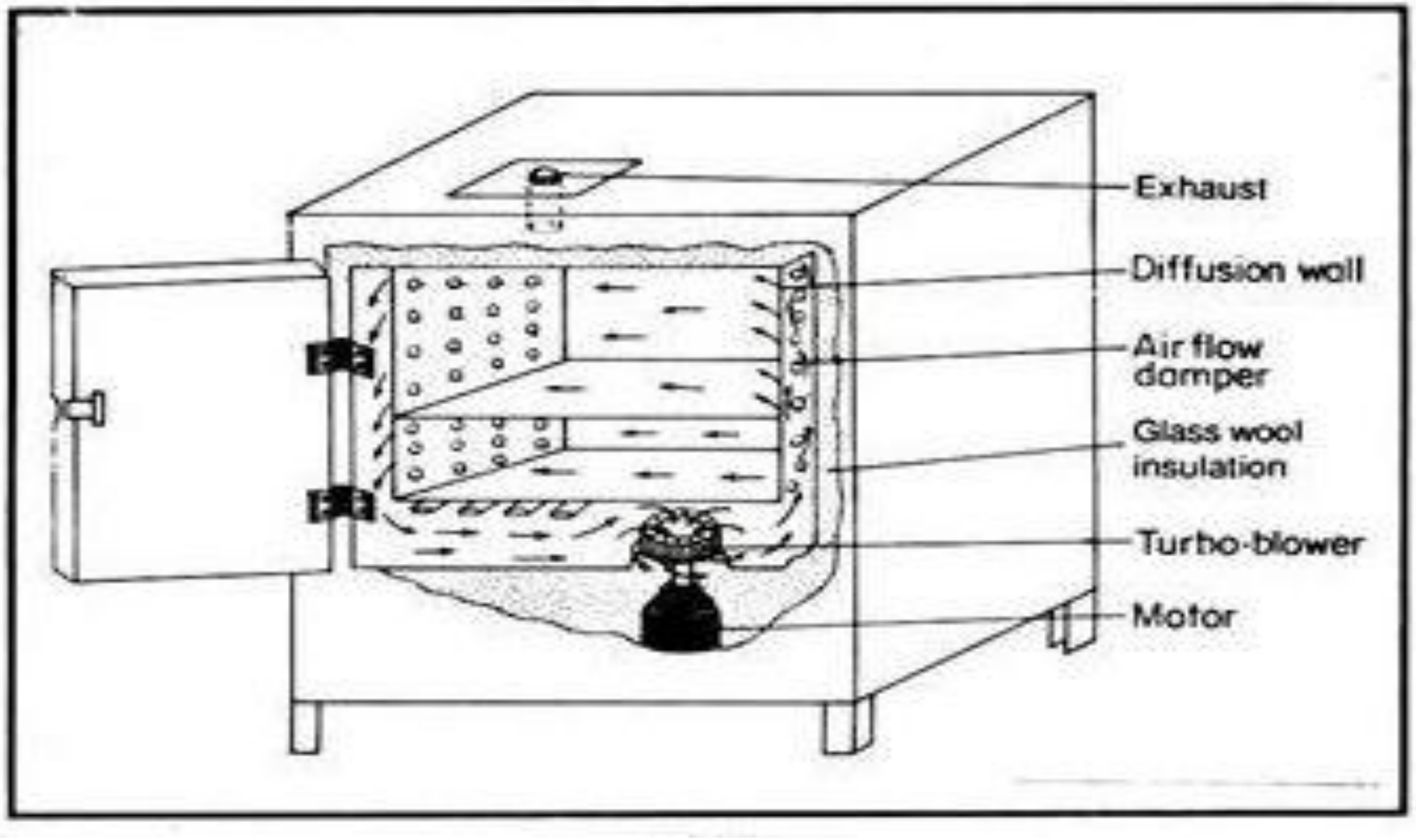
HOT AIR OVEN

- Containers must be placed on their side for the same reason to avoid impeding of hot air.
- Powders , oils, fats & grease must be packed in small quantites or in thin layers to enable penetration o heat.

Uses: Sterilization of glassware(test tubes, flasks, pipettes, syringes,petridishes)

- Sterilization of sharp surgical instruments (forceps,scalpels,scissors)
- Sterilization of powders,fats,grease,that are not penetrate by moist heat.

HOT AIR OVEN


Exhaust
Diffusion wall
Air flow damper
Glass wool insulation
Turbo-blower
Motor

MOIST HEAT :-

- Most efficient means of sterilization.
- Acts by denaturation and coagulation of proteins .

3 RANGES OF TEMP USING MOIST HEAT :-

- Below 100℃ are not considered as sterilisation as spores of
- At 100℃ some acteria are not destroyed.
- Above 100℃

MOIST HEAT :-

BELOW 100 ˚C

a) Pasteurization :-

Principle : process of heating a liquid to a specific temperature for a definite length of time and then cooling it immediately.

Uses : Milk processing.

I. HOLDER METHOD : heated at 63 ˚C for 30 min.

II. FLASH METHOD : heated at 72 ˚C for 15-20 min.

b) Water bath :-

- For vaccines of non – sporing bacteria – 60 ˚C
- For serum or protein – 56 ˚C/ hr for sever successive days.
- For cystoscopes, specula – 75 ˚C / 10 min .
- Clothing , bed clothes , eating utensils , nursing equipment – washing at 70-80 ˚C for several mins .

c) Inspisation :-

- Used for the sterilisation of culture media that contain heat sensitive ingrediants.
- The temperature used is 80-85 ℃ for 1\2 hour on successive days.Eg:lawnstein jenson media,loeffer serum slope.
- The purpose of doing on 3 successive days is that on the first day vegetative bacteria are killed.
- On the 2^{nd} day, germinated spores are killed and on the 3^{rd} day any other organism present is killed.

About 100 ̊C:-

a)Boiling :-

- Kills vegetative bacteria at 90-100 ̊C but sporing bacteria require a longer time. Surgical apparatus disinfected by boiling for atleast 30 minutes.

B)Steam at 100 ̊C:- (at atmospheric pressure)

- Tyndallization : It consists of exposure at 100 ̊C for 20 mins on 3 successive days. It is used for media containing sugars & gelatin. This process is also known as FRACTIONAL STERILIZATION. The instrument used is the koch’s or Arnold’s steamer.

Above 100 ̊C:-

- **Autoclave:** The instrument used for the sterilisation is called Autoclave or steam steriliser.

- **Principle:**
- H2o boils when its vapour pressure equals that of the surrounding atmosphere.
- Hence when pressure inside a closed vessels, increases the temperature at which H2O boils also increase producing dry saturation steam.
- .

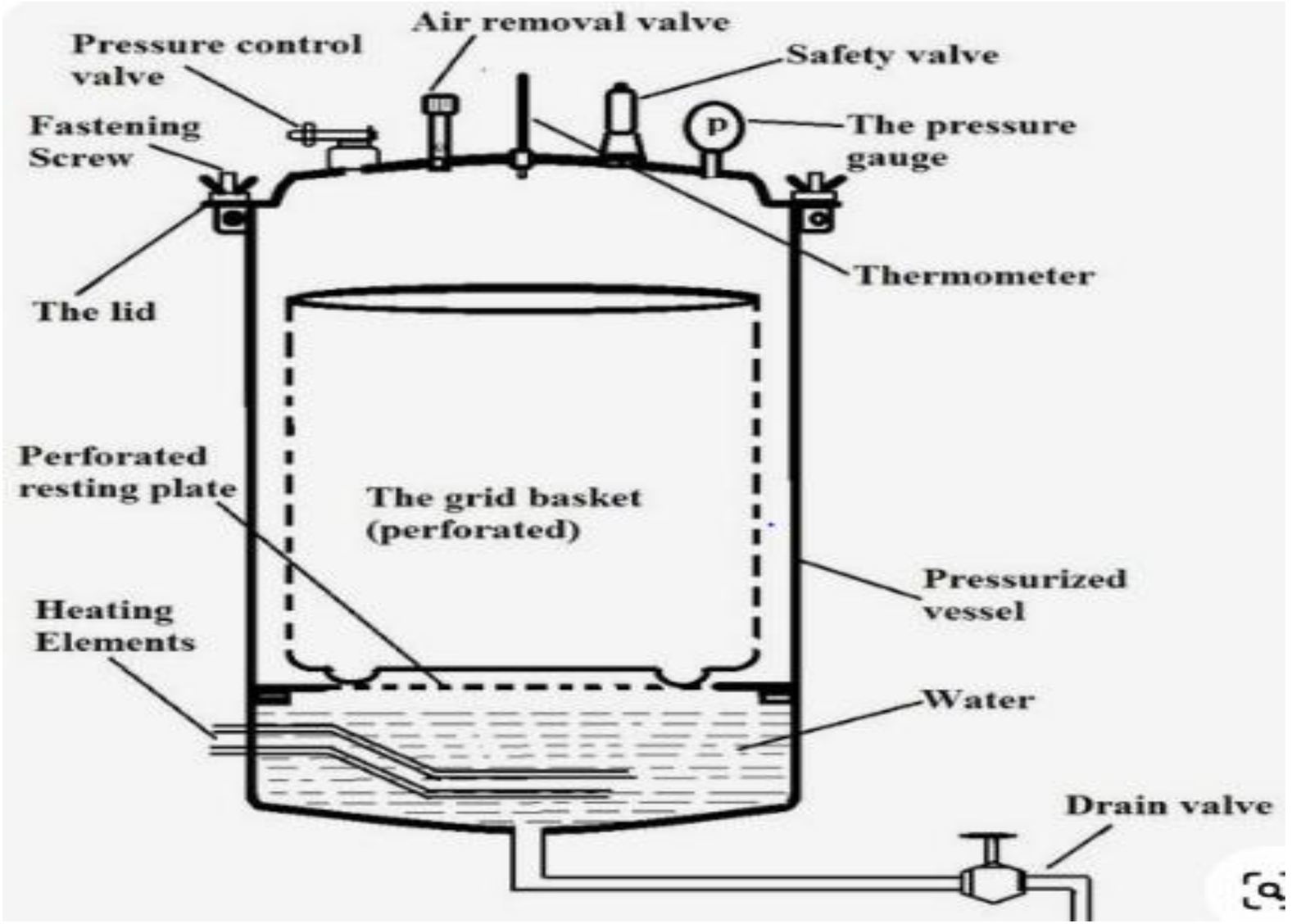
Principle:

- Approximately 80% of the latent energy is dry saturation steam is in the form of latent heat. That is released when the touch a cooles object and condenses.
- Condensation is accompanied by an instantaneous contraction of the stream (1600 ml steam at 100℃ condenses to 1ml) creating low pressure region in which more steam flows.
- This results in penetration of steam into articles being sterilized

Types :-

1) Simple laboratory autoclave
2) Steam jacketed autoclave
3) Prevaccum sterilization

STRUCTURE AND PROCESS :-



❖The basicstructure consists of a ventrical or horizontal consists made of gunmental or stainless stell with a supporting frame . It has the following features:

❖A lid or door fastened by screw clamps and made airlight with a suitable washes.

❖ Pressure gauge.

❖Discharge tap on lid to allow air to escape.

❖ Safety valve that can be set to go off at the required pressure.

❖Thermometer / Temoperature gauge

❖Heating by gas or electricity.

❖The articles to be sterilized are placed on a perforation tray and should not be packed closely.

❖The lid is closed high & the autoclave is heated.

❖The discharge tap is kept open so that air stream mixture can escape.

❖Once all air is discharged, the discharge tap is closed.

❖The pressure of steam inside the autoclave rises and when the desired pressure is reached i.e,15 pounds/sq.inch the safety valve opens and excess steam escapes.

❖The process of sterilization is now kept going for 15-20 mins after the required pressure reached. The temperature attained is 121 ℃.
❖After the period of sterilization is completes the autoclaves is switched off and allowed to cool sufficiently till pressure, inside equal to the atmosphere pressure.
❖It should not be opened earlier as there may be spilling of liquids or an explosion.

uses:-

❖Sterilization of culture media & glasswares .
❖Sterilization of blunt surgical instruments and OT clothes.
❖Destruction of infected materials like blood, sputum.

RADIATION

- NON IONISING
- IONISING

- **NON IONISING RADIATION**:
- Infra red rays are used for mass sterlization of syringes & catheter.
- The temperature attained is 190˚C for 6mins or 180˚C for 12 mins
- UV radiation in the range of 240-280 are used for sterlization but 260nm in the considered bactericidal.
- They act by formation of pyramidine dimus on DNA strand.
- It is used to sterilize air and surface in room and biosafety cabinets.
- Its is used for the purification of water.

RADIATION

- **IONISING RADIATIONS**:

- They include x rays, gamma rays and high speed elections
- They cause damage to DNA
- They are used to sterilize antibiotic, hormones, sutures ,plastics,syringes and catheters.
- As there is no increase in the temperature of the article being sterilized it is also referred to as cold sterilization.

FILTRATION:

- CANDLE FILTER
- ASBESTOS FILTER
- SINTERED GLASS FILTER
- MEMBRANE FILTER

In this process, bacteria are removed from heat sensitives such as serum, sugar, solutions or antibiotics used for culture media preparation.

CANDLE FILTER :-

- They are made of diatomaceous earth or kise lguhr and are available in different grades of pores sizes .
- They are used for purification of water for industrial and drinking purpose .

THE GRADES AVAILABLE ARE :

- V (viel) - coarsest
- W (wening) – finest
- N (normal) – intermediate

EG : Birk feld , mandles , chamberland .

ASBESTORS FILTERS :-

- They are disposable single use filters made of magnesium silicate .
- EG: Seity & Sterimat filter

SINTERED GLASS FILTER :-

- They consists of finely ground glass piecess that are flused together and are available in different pour sizes.

MEMBRANE FILTER :-

- They are porous membrane about 0.1 mm thick made of cellular acetate, cellulose nitrate polycarbonate and polyvinylinder fluoxidereor some other synthetic material.
- The membrane are supported on a frame and mild in spicial holders .
- Fluids are made to transverse membrane by positive or negative pressure by centrifugation.

HEPA FILTER :-

- They are high efficiency particulate air filterused to deliver bacteria free air into room.
- They are usedin operation rooms & burn units to remove bacteria from air .
- They are used also used in laminar air flow system to prevent infestion

THANK YOU