

STERILIZATION

Sterilization is the process of killing or removing bacteria and all other forms of living organism and their spores from preparation or articles.

Method of Sterilization

Method of sterilization have been divided into following categorie



1) Physical Methods

- Dry heat sterilization
- Moist heat sterilization
- Sterilization by radiations

2) Chemical Methods

- Gaseous sterilization
- Sterilization by disinfectants

3) Mechanical Methods

- Sterilization by filtration

Physical Methods

a) Dry Heat Sterilization

Substances which are destroyed by moist heat may be sterilized by moist heat. Dry heat can be used to sterilize items but as the heat takes much longer to be transferred to the organism both the time and the temperature must usually be increased unless forced ventilation of the hot air is used. The standard setting for a hot air oven is at least two hours at 160 °C (320 °F).

A rapid method heats air to 190 °C (374 °F) for 6 minutes for unwrapped objects and 12 minutes for wrapped objects. Dry heat has the advantage that it can be used on powders and other heat-stable items that are adversely affected by steam.

Substances Sterilized:

Substances which are sterilized by dry heat include fixed oils, liquid paraffin, petroleum, propylene glycol and powders. In addition to this, it is also applied to sterilize glassware, many surgical instruments and surgical catgut. Volatile preparations or substances and surgical dressings cannot be sterilized by this method.

Mechanism:

During dry heat sterilization, the microorganism and bacteria spores are killed by oxidation since dry heat is less effective than moist heat. Higher temperature and longer period of exposure are required. Exposure at 160 degrees for one hour is required for dry heat sterilization.

Advantages

- Suitable for substances destroyed by moisture
- Glass wares like flasks, test tubes, pipettes can be sterilized
- Less damaging to glass and metal equipments than moist

Disadvantages

- Cannot be used for volatile and thermolabile substances.
- Required long heating time and high temperature
- Not suitable for surgical dressings.

Methods**1. Flaming**



It is simplest method of dry heat sterilization in which the material to be sterilized is kept in the hot part of the Bunsen burner flame for few seconds and the process is repeated several times. This method is generally used for those articles which are to be used immediately for example forceps, blades, knives, needles, wire loops, metal spatulas.

2. Hot Air Oven

It consists of a metallic chamber of aluminium or stainless steel, which is electrically heated and thermostatically controlled. There are two types.



- i. In which air is circulated by gravity convection to all parts of the chamber.
- ii. Mechanical convection type in which air is circulated by fan. The latter type is more satisfactory because sterilizing temperature is controlled.

Glassware, conical flasks, test tubes, pipettes etc. are sterilized by this method. They should be plugged with non-absorbent cotton wool because absorbent cotton wool becomes saturated during the process.



3. Incineration

Incineration will also burn any organism to ash. It is used to sanitize medical and other biohazardous waste before it is discarded with non-hazardous waste.



b) Moist heat sterilization

It is the most reliable method of sterilization because in the presence of moisture bacteria are destroyed at a considerably lower temperature rather than dry heating.

Mechanism

By this method the microorganisms are destroyed by denaturing and coagulation of some of the essential proteins present in the microorganisms.

Advantages

- Microbes are killed more effectively
- Ampoules are readily sterilized by this method
- Bulk quantities surgical dressing and surgical instrument are effectively sterilized

Disadvantages

- Thermolabile substances and ointments can not be sterilized.

Method

- i. Autoclaving
- ii. Heating with bactericide
- iii. Heating with boiling water
- iv. Tantalization

1. Autoclaving

Autoclaving is the process of heating in an autoclave in which saturated steam under pressure is allowed to penetrate through the material for 20 minutes at temperature of 121°C.

Autoclave

It is an apparatus used for sterilization by **steam under pressure**.

**Working**

- ✓ Autoclaves commonly use steam heated to 121 C or 134C.
- ✓ To achieve sterility, a holding time of at least 15-20 minutes at 121 C or 3 minutes at 134 C is required.
- ✓ Additional sterilizing time is required for liquid and instruments packed in layer of cloth as they may take longer to reach the require temperature.
- ✓ Proper autoclave treatment will in activate all fungi, bacteria, viruses and also bacterial spores which can be quite resistant.
- ✓ It will no necessarily eliminate are prions.
- ✓ For prion elimination, various recommendation states 121-132C for 60 minute or 134C for at least 18 minutes.

Precautions:

- ✓ For effective sterilization steam needs to penetrate the autoclave load uniformly.
- ✓ During the initial heating of the chamber, residual air must be removed.
- ✓ For autoclaving, as for all disinfection of sterilization methods cleaning is critical.
- ✓ Cleaning can also removed large number of organisms.
- ✓ Cleaning instruments or utensils inorganic matters, cool water must be used.
- ✓ Treatment with ultrasound or pulsed air can also be used to removed debris.

2. Heating With A Bactericide

- ✓ In this method bactericide is added to the solutions to be sterilized which are the sealed.
- ✓ The sealed containers are then heated at 100C for 30 minutes in water bath.
- ✓ Commonly used bactericide includes benzalkonium chloride, chlorocresol.

3. Sterilization by boiling water

- ✓ The boiling water bath is most use full for sterilizing instruments like syringes, knives, blades, scissors and others. They are completely dipped in boiling water for 20 minutes.

4. Tyndallization

Tyndallization named after John Tyndall is a lengthy process designed to reduce the level of activity of sporulating bacteria that are left by a simple boiling water method.

- ✓ The process involves boiling for a period at atmospheric pressure, cooling, incubating for a day and finally boiling again.
- ✓ The three incubation periods are to allow heat-resistant spores surviving the previous boiling period to germinate to form the heat sensitive vegetative stage which can be killed by next boiling steps.
- ✓ Tyndallization ineffective against prions.



c) Sterilization by Radiation

Sterilization by radiation is also known as cooled sterilization because no heat is used in this method. The microorganisms are very susceptible to lethal effects of radiation.

Mechanism

- ✓ By radiations, alteration of chemicals takes place present in microorganisms with the formation of new compounds which destroy the microbes.





The vital structures of cells such as nucleoproteins are destroyed by radiations which kill the microbes.

Advantages



Used in the preservation of food and parenterals containing antibiotics.



Used for the sterilization of some bacterial and viral vaccines.



No aseptic handling is required because sterilization can be done after packing.

Disadvantages



High cost.



Radiations are harmful to the persons operating.



Radiations may lead to change in color, texture and solubility.

Methods

Methods exist to sterilize using radiation such as electron beams, X- rays, gamma rays, or subatomic particles.

Gamma rays



Gamma rays are very penetrating and are commonly used for sterilization of disposable medical equipment, such as syringes, needles, cannulas and IV sets.

Electron beam processing



Electron beam processing is also commonly used for medical device sterilization, Electron beams use an on-off technology and provide a much higher dosing rate than gamma or x-rays.



Due to the higher dose rate, less exposure time is needed and thereby any potential degradation to polymers is reduced.



A limitation is that electron beams are less penetrating than either gamma or x-rays.

X-Rays



X-rays are less penetrating than gamma rays and tend to require longer exposure times, but require less shielding, and are generated by an x-ray machine that can be turned off for servicing and when not in use.





Ultraviolet light

- ✓ Ultraviolet light irradiation (UV, from a germicidal lamp) is useful only for sterilization of surfaces and some transparent objects.
- ✓ Many objects that are transparent to visible light absorb UV.
- ✓ It also damages many plastics, such as polystyrene foam.

2 Chemical Sterilization

- ✓ Chemicals are also used for sterilization. Although heating provides the most reliable way to rid objects of all transmissible agents, it is not always appropriate, because it will damage heat-sensitive materials such as biological materials, fiber optics, electronics, and many plastics.

Chemical sterilization includes:

1. Ethylene oxide
2. Ozone
3. Chlorine bleach
4. Glutaraldehyde
5. Formaldehyde
6. Hydrogen peroxide
7. Peracetic acid
8. Prions

Chlorine bleach

- Chlorine bleach is another accepted liquid sterilizing agent.



- House hold bleach consists of 5.25% Sodium hypochlorite.
- It is usually diluted to 1/10 immediately before use; however to kill *Mycobacterium tuberculosis*. It should be diluted only 1/5.
- The dilution factor must take into account the volume of any liquid waste that it is being used to sterilized.
- Bleach will kill many organisms immediately, but for full sterilization it should be allowed to react for 20 min.
- Bleach will kill many, but not all spores.
- It is highly corrosive and may corrode even stainless steel, surgical instruments.

Ozone

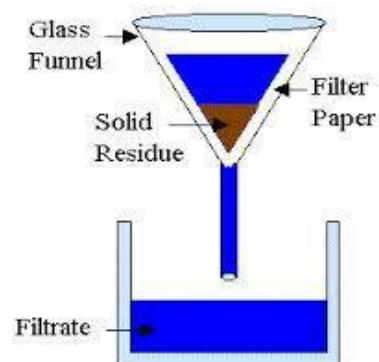
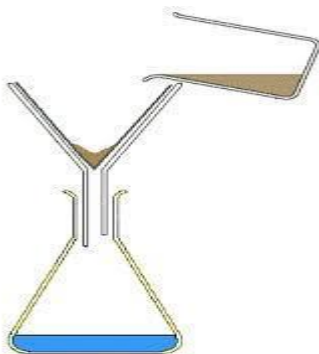
Ozone is used in industrial setting to sterilize water and air, as well as a disinfectant for surfaces.

It has the benefit of being able to oxidize most organic matter. On the other hand, it is a toxic and unstable gas that must be produced on sight, so it is not practical to use in may settings.

3. Mechanical methods

It includes filtration.

Filtration



Involves the physical removal (exclusion) of all cells in a liquid or gas. It is especially important for sterilization of solutions which would be denatured by heat (e.g. antibiotics, injectable drugs, amino acids, vitamins, etc.) portable units can be used in the field for water purification and industrial units can be used to “pasteurize” beverages. Essentially, solutions or gasses are passed through a filter of sufficient pore diameter to remove the smallest known bacterial cells.

Preservatives:

Static agents used to inhibit the growth of microorganisms, most often in foods. If eaten they should be nontoxic and sulfur dioxide.

Antimicrobial agents

Are chemicals that kill or inhibit the growth of microorganisms.



Antimicrobial agents include chemical preservatives and antiseptics, as well as drugs used in the treatment of infectious diseases of plants and animals.



Antimicrobial agents may be of natural or synthetic origin, and they may have a static or cidal effect on microorganisms.

Types of antimicrobial agents**Antiseptics:**

Microbial agents harmless enough to be applied to the skin and mucous membrane; should not be taken internally.

Examples

Include alcohols, mercurial, and silver nitrate, and iodine solution, alcohols, detergents.

Disinfectants:

Agents that kill microorganisms but not necessarily their spores but are not safe for application to living tissues, they are used on inanimate objects such as tables, floors, utensils etc.

Example

Includes hypochlorite chlorine compound dye, copper sulfate, quaternary ammonium compounds, and formaldehyde and phenolic compounds.

Application of sterilization processes

The pharmacist uses sterilization process in pharmaceutical tries for sterilization of glassware and other equipments; the preparation of injectables, ophthalmic, irrigating and sterile dosage forms ; and for the dilution of sterile medicaments. However, it must be kept in mind that all types of medicaments and other articles cannot be sterilized by any one method; the pharmacist should have thorough knowledge of sterilization processes that for certain kind of medicament or article, whether dry heat, moist heat, filtration method or any other method is to be used.

In hospital practice the sterilization processes are used for the preparation of sterile gauzes and dressings; for sterilization of glass equipment like test tubes, petri dishes, pipettes, all glass syringes etc. for sterilization of operation theatre, and all types of articles used in operation theatre including aprons, gloves, face masks, hoods, forceps, scissors, knives etc.

The following articles/medicaments require sterilization:

1) Glassware

The glassware and apparatus like flasks, beakers, funnels, dropper bottles, glass rods, pipettes, petridishes, glass tube pestle & mortar, tiles, ointment tubes, ampoules, vials syringes, needles etc. may be sterilized either by moist or dry heat but dry heat method is preferred. .

2) Equipment

Apparatus or devices made up of metal and surgical instruments, spatulas, metal part of bacterial filters such as millipore filter, Seitz filter etc may be sterilized by steam under pressure at a temperature of 121°C for 15 to 30 minutes, depending on the size of instruments. The materials should be wrapped with muslin so that steam may penetrate to the surface of the metal parts. Sharp edged instruments should not be sterilized in hot air because long heating at high temperatures in hot air lead to oxidation which reduces the sharpness of the blades.

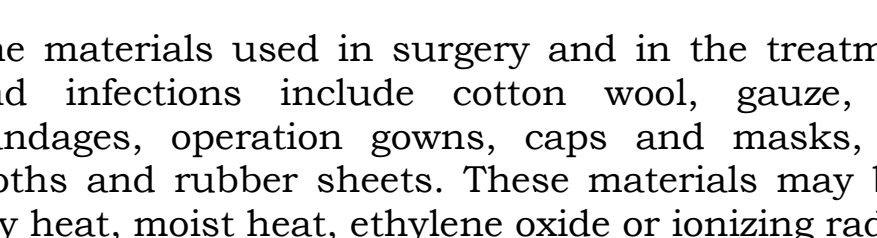
(3) Rubber and plastics

Rubber articles such as stoppers, gloves, certain catheters and special feeding tubes may be sterilized in the autoclave at 121°C

for 20 minutes they must not be subjected to dry heat because high temperatures will spoil the rubber articles. Some synthetic rubbers such as silicon rubber has good heat resistance power therefore may be sterilized either by dry heat or moist heat.

**(4) Aqueous/non-aqueous solutions and suspensions**

Thermostable aqueous solutions should be sterilized by steam under pressure in an autoclave at a temperature of 115°C to 118°C for 30 minutes but medicaments which are sufficiently thermostable may be sterilized at a temperature of 121°C for 15



The materials used in surgery and in the treatment of wounds and infections include cotton wool, gauze, gauze swabs, bandages, operation gowns, caps and masks, towels, trolley cloths and rubber sheets. These materials may be sterilized by dry heat, moist heat, ethylene oxide or ionizing radiations.