STERILIZATION & DISINFECTION

By

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Decontamination

- Any procedure that reduces pathogenic microbes to a level where items are safe for handling & disposal.
- It can be achieved by **cleaning, disinfection and sterilization.**

Cleaning

- A process that removes foreign material (dirt, organic matter).
- Must precede disinfection and sterilization.
- **Usually done with soap and water or detergent.**
Disinfection

- Destruction of most but not necessarily all pathogenic microbes or their *spores*.

Sterilization

- Killing of all living forms of microbes including spores.
Antiseptics

- Chemical compounds that could be applied topically on **animate surfaces**.

Disinfectants

- Chemical compounds applied for **inanimate surfaces**.
Importance of sterilization and disinfection:

- Safety in the laboratory.
- The patient safety depends on using proper methods of sterilization to prepare instruments, needles, IV fluids.
- The accuracy and validity of microbiological tests.
DISINFECTION

- **Heat**
  1. Moist heat at temperature below 100 degree (Pasteurization).
  2. Moist heat at temperature of 100 degree
    - Boiling
    - Steaming

- **Radiation**: (Ultraviolet rays)

- **Chemical**: (Disinfectants)
Disinfection by heat

1. Moist heat at temperature below 100 degree
   - Used for milk disinfection.
   - Milk is heated either at 63 degree for 30 minutes or 72 degree for 20 seconds and immediately cooled to below 10 degree.
2. Moist heat at temperature 100 degree:

- Heating at 100 degree for 20 minutes.
- Used for disinfection of surgical and medical equipments in emergency.
Done in Koch’s sterilizer.

This sterilizer is vertical metal cylinder with removable conical lid having a small opening for escaping steam.

The articles to be sterilized are placed on a perforated tray situated above water which is placed in the bottom of the cylinder.
(Tyndallization)

- Intermittent sterilization by exposure to steam at 100 degree for 20-45 min. for three successive days.
- Used for sterilization of sugar media which decompose at high temperatures.
- The principle is that one exposure will kill only vegetative bacteria. Between heatings, the spores will vegetate to be killed during subsequent exposure.
Disinfection by radiation

Ultraviolet rays

- Present in sun rays or artificially produced by mercury lamp have weak penetrating power.
- Used to reduce the number of bacteria in air inside operation rooms, laboratory safety cabinet.
Disinfection by chemical disinfectants

- Disinfectants may be:
  - High level disinfectants.
  - Intermediate level disinfectants.
  - Low level disinfectants.
High level disinfection
- **large number of spores after prolonged exposure**
- Vegetative bacteria
- *Tubercle bacilli*
- Fungi
- Viruses

Intermediate level disinfection
- **Few number of spores**
- Vegetative bacteria
- *Tubercle bacilli*
- Fungi
- Enveloped viruses (HBV, HIV)

Low level disinfection
- **Mainly vegetative bacteria**
- Some fungi
- Narrow range of viruses
Organisms according to the innate resistance

1. Prions (the most resistant)
2. Spores
3. Tubercle bacilli
4. Non enveloped viruses
5. Fungi
6. Vegetative bacteria
7. Enveloped viruses such as HBV, HIV
1 Alcohol

- Ethyl alcohol or isopropyl alcohol 70%.
- Used as skin antiseptic.
- Intermediate level disinfectant.
- Exposure for at least 5 minutes is needed to achieve adequate disinfection.
2. Glutaraldehyde

- Available in 2% concentration (cidex).
- High to intermediate level disinfectant.
- Disinfection of instruments that can not withstand heat such as endoscopes.
Chlorine releasing compounds

- Example of chlorine releasing compounds is:
  - Hypochlorite solution (such as household bleach)
  - Intermediate level disinfectant.
  - Widely used in homes, hospitals and laboratories to disinfect table tops, incubators, spilled cultures.
  - Disinfection of water supply.
Iodophores

- Examples of iodophores:
  - Tincture iodine (2% iodine in ethanol)
  - Betadine (10% povidone-iodine)
- Intermediate to low level disinfectant.
- Used for disinfection of surgical wounds.
Example:

- Dettol, lysol (2% concentration)
- Intermediate level disinfectant.
- Used in laboratories to disinfect spilled cultures on working areas or in discard jars.
Quaternary ammonium compounds

- Example:
  - ✓ Cetavlon, savlon
  - Low level disinfectant.
  - Used to clean floors, walls.
  - Inactivated by organic matter.
  - Not affect Gram negative bacilli.
Hydrogen peroxide

- Available as 3% H2O2
- High to intermediate level disinfectant.
- Used as antiseptic for wounds, disinfectant for contact lenses.
8. Heavy metals
   - As mercury in mercurochrome and silver in silver nitrate.

9. Chlorhexidine
   - Safe antiseptic used for hygienic and surgical hand washing and for oral hygiene.
STERILIZATION

- **Chemical methods (Cold sterilization)**
- **Heat**
  1. Dry heat (Red heat, hot air oven, incineration)
  2. Moist heat at temperature above 100 degree (Autoclave)
- **Radiation**: (Ionizing radiation & infrared rays)
- **Filtration**: (Fluid filters & air filters)
- **Gaseous sterilization**: (Ethylene oxide + gas plasma)
Chemical sterilization

- Glutaraldehyde (contact time 10 hours).
- Liquid peracetic acid.
- H2O2 6%.
Sterilization by heat

Dry heat

Moist heat
Less effective than the moist heat.

Examples of dry heat are:

1) **Red heat:**

   Sterilization of the bacteriological loop by heating in the Bunsen flame or electric incinerator till becomes hot red.
2) **Hot air oven:**
   ✓ Temperature of 160 degree for 2 h. or 170 degree for 1 h.
   ✓ For glass ware, and metallic instruments.

3) **Incineration:**
   ✓ Destruction of contaminated materials in the incinerator.
1) **Principle:**

- When water is heated in a closed vessel under pressure, the boiling point of water rises above 100 degree.
- Water is heated at 2 atmospheric pressure and the boiling temperature will be 121 degree or at 3 atmospheric pressure and the boiling temperature will be 134 degree.
2) **Autoclaving is highly efficient because:**

- High temperature.
- High penetrating power of the steam under pressure.
- When steam condenses on the articles, it liberates latent heat to the articles to be sterilized.
- Non toxic
- Not time consuming.
Simple laboratory autoclave
3) **Directions for use of the simple laboratory autoclave:**

- It is a metal cylinder with tightly fitting lid. The lid is connected to a steam discharge tap, safety valve and manometer.

- Water is placed in the bottom and the articles to be sterilized are placed on top of a perforated tray above water level, then the lid is tightly closed.

- Water is heated electrically.
The steam discharge tap is opened and the safety valve is adjusted to 15 ib per square inch = double atmospheric pressure.

Steam will come out of the tap in interrupted jets as it mixes with air.

When steam is released in a continuous stream, the tap is closed.

When the steam pressure reaches the desired level, safety valve will allow excess steam to escape.
From this point, the holding sterilization time which is 20-30 minutes is calculated.

Then, the heater is turned off and the autoclave is allowed to cool down before opening the lid.

4) **Autoclave is used for sterilization of:**

- Surgical instruments and dressings.
- Bed linen.
- Cotton, gauze
- Culture media not destroyed by heat.
5) **Steam jacketed autoclave:**

- Double walled chamber in which steam at 121 degree is introduced from an external source.
- Steam enters the jacket from which it enters the chamber migrating downwards replacing the air which is expelled through a discharge tap at the bottom.

6) **Flash autoclaves:**

- It sterilizes at 134 degree for 3 – 5 minutes.
- Used in operation rooms in emergency situations.
7) **Testing the efficiency of autoclave:**

- **Mechanical methods:**
  - Daily measurement of temperature, pressure gauge.

- **Chemical indicators:**
  - Change its color at the end of sterilization cycle.

- **Biological indicators:**
  - Using spores of *Bacillus stearothermophilus*.
  - However, spores of *Bacillus subtilis* are used to test the efficiency of hot air oven.
Ionizing radiation:
- Such as gamma rays emitted from radioactive cobalt 60 or beta rays emitted from electron accelerators.
- High penetrating power.
- Used for plastic syringes, catheters, gloves, surgical sutures.
Sterilization by filtration

Fluid filters
- Used for sterilization of biological fluids destroyed by heat such as serum, plasma, vitamins, hormones.

Air filters
- Air is filtered by HEPA filters.
- HEPA means high efficiency particulate air way arresters.
- Air filtration is needed in operation rooms, safety cabinets.
1) **Vacuum or Seitz filter:**

- Formed of asbestos disc which is inserted into a metal holder connected to a flask.
2) **Millipore (membrane) filters:**

- Synthetic membranes made from cellulose diacetate.

3) **Syringe filter:**

- Membrane filters 13 – 25 mm in diameter in a small holder connected to a syringe containing the fluid to be filtered.
**Sterilization by gases**

- **Ethylene oxide gas**
  - Used for plastic and rubber articles.
  - Ethylene oxide gas is toxic, explosive and carcinogenic to laboratory animals.

- **Plasma gas sterilizer**
  - Plasma means any gas which is formed of ions, electrons, neutral particles.
  - Used for surgical instruments mainly those with narrow lumen such as arthroscopes & laparoscopes.
  - Non toxic.
### Selection of adequate level of contamination

<table>
<thead>
<tr>
<th>Item</th>
<th>Characteristics</th>
<th>Example</th>
<th>methods</th>
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<tbody>
<tr>
<td><strong>Critical</strong></td>
<td>Instruments that enter sterile tissues, cavities or vascular system</td>
<td>Surgical instruments, needles, catheters</td>
<td>Sterilization by autoclave, plasma gas sterilizer or ethylene oxide gas</td>
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<tr>
<td><strong>Semi-critical</strong></td>
<td>Objects that come in contact with non intact skin or mucous membranes</td>
<td>Endoscopes, thermometers</td>
<td>High level disinfection</td>
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<tr>
<td><strong>Non-critical</strong></td>
<td>Objects that come in contact with intact skin</td>
<td>Sphygmomanometer, bed linen</td>
<td>Intermediate to low level disinfection</td>
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