Sterilization & Disinfection

* Fighting Bacteria acc to it's site: → inside the Body ⇝ use Antibiotics.
→ outside the Body ⇝ use sterilization & disinfection methods.

* Definitions:

*** Sterilization:** Killing all forms of microorganisms including Bacterial spores By physical & chemical methods.

↳ physical methods

1. Heat (moist, dry)
2. Radiation
3. Filtration

↳ Chemical methods

1. Gaseous & liquid Chemicals

↳ uses of Stirilization:

1. Sterilization of surgical instruments.
2. Sterilization of syringes.
3. Sterilization of gloves.
4. Sterilization of Catheters.
5. Sterilization of Culture media.

*** Disinfection:** Killing most (if not all) forms of microorganisms Except Bacterial spores By physical & chemical methods.

↳ physical methods

1. Moist heat

↳ below 100°C (Pasteurization)

* At 63°C for 30 min.
or * At 72C for 20 sec.
* Should be followed by rapid cooling to Not make a favourable environment for thermophilic Bacteria.
* Kills: - mycobacterium tuberculosis
- Brucilla abortus → cause Brucillosis.
- Salmonella → cause typhoid fever.
- Coxiella burnetti → cause Q-fever.

↳ At 100°C for 20 min (Boiling)

* Kills all vegetative Bacteria
* used in emergency (when No other method available)
* used for disinfection of: - Glass syringes & surgical instruments.

2. Radiation

* Ultraviolet rays (low penetration)
* Artificially from Mercury lamps
* Advantage: act as Bactericidal (kills Bacteria)
* Disadvantage: Carcinogen (causes cancer)
↳ that's why should be used in empty room.

* used in:
↳ operation room
↳ surface disinfectant
↳ Drug filling cubicles
↳ Safety cabinets

↳ Chemical methods

1. Disinfectants: Chemical Substances that are used to acheive Disinfection. (They are Toxic) (high, intermediate, low levels)

Chemical Methods of Disinfection

* using Disinfectants of diff levels (high, intermediate, low)

* they have Combination of actions on microbs including:
  1. Oxidation
  2. Denaturation
  3. Cell wall & cell membrane damage
  4. Breaks DNA

  ↳ that's why usually there is No resistant for disinfectants, opposite of Antibiotics that work on one target & there is resistant for it.

* levels of Disinfectants:

A. low level Disinfectants: Kill most microbs Except Bacterial spores & Mycobacterium tuberculosis.

↳ examples:

1. Quaternary ammonium Compounds: - Benzethonium Chloride
   - Benzalkonium Chloride

↳ used in:

1. disinfection of floors & Blood spills.

B. Intermediate level Disinfectants: Kill most microbs Except Bacterial spores.

↳ examples:

1. Alcohols 70% ⇒
   * mechanism of Killing:
     ↳ Denaturation
     ↳ Membrane damage
     ↳ Disruption of lipid containing
   * Act as Bactericidal, virucidal, Fungicidal
   * examples:
     ↳ Ethanol & Isopropanol ⇒ used as: Antiseptic & hand sanitizers
     ↳ Methanol ⇒ very dangerous, Cause Blindness, damage in Brain, death

2. Phenols ⇒ (↳ first used in operation rooms by Lister)
   * mechanism of Killing:
     ↳ Denaturation
     ↳ Membrane damage
   * examples:
     ↳ Phenol derivatives: - cresol (lysol)
     - Chloroxylenol
   * used in:
     ↳ disinfection of floors & culture spills.

3. Biguanides ⇒
   * examples:
     ↳ Chlorohexidine
   * used in
     ↳ mouth washing

4. Halogenes ⇒
   * mechanism of Killing:
     ↳ Denaturation
     ↳ Oxidation
   * examples:
     ↳ Iodines: - Tincture iodine (2% iodine + 2.4% sodium iodide in 50% ethanol)
     - Betadine (Povidine + iodine)
     * Both used as Skin antiseptics but Betadine Stronger.

     ↳ Floride * in tooth paste

5. Heavy Metals:

* mechanism of Killing:
  - ↳ Denaturation
  - ↳ Inhibition of enzymatic activity

* examples
  - ↳ Zinc, Copper, Nickle, Silver (Zn, Cu, Ni, Ag)

* Informations:
  - ↳ They have Antimicrobial activity.
  - ↳ Cu, Zn, Ni used to make doorknobs to reduce contamination in hospitals.
  - ↳ water can be stored in Silver jugs for drinking.
  - ↳ They are Toxic to humans & animal in excessive concentrations (↑ conc.)
    - ↳ can cause Argyria (Bluish-gray skin)
  - ↳ Silver Nitrate Drops: given to infants with Ophthalmia Neonatorum
    - ↳ if pregnant woman had "gonorrhea" caused by "Neisseria gonorrhoeae" bacteria → the infection will be transmitted to the baby causing Ophthalmia neonatorum.)
  - ↳ Zinc oxide: found in Calamine lotion & Baby powder.

C. High level Disinfectants: Kill most microbs Except large amount Bacterial spores. → can kill ↓ amount Bacterial spores
↳ act as Sterilant in this case

↳ examples:

1. Chlorine
   - ↳ disinfectant for water & swimming pools.

2. Sodium hypoChlorite (Sodium + Chlorine + oxygen)
   - ↳ disinfectant in homes & hospitals, But it's Corrosive
     - ↳ *cause decolorization if put with Clothes
     - * if used with metals it can cause corrosion

3. Hydrogen peroxide (H_2O_2)
   - ↳ Antiseptic
   - ↳ disinfectant for contact lens.

4. GlutarAldehyde 2% & Peracitic Acid
   - ↳ Need to be used for ~10 hours to get high level disinfection.

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* Antiseptics: Killing most (if not all) forms of microorganisms Except Bacterial spores with Non-toxic materials (can be used on living tissue)
  - ↳ same as disinfection with difference that antiseptic is Non-toxic while disinfectants are toxic.

* Germicide: Agent that destroy microorganisms, and it's name according to where it's used & what it kills:
  - ↳ Bactricide: Kills Bacteria.
  - ↳ Virucide: Kills viruses.
  - ↳ Fungicide: Kills fungi.

* Can act as:
  - → Sterilant if acheived sterilization (Killing including Bacterial spores)
  - → Disinfectant if acheived disinfection (Killing Except Bacterial spores)
  - → Antiseptic if acheived disinfection but it's Not Toxic.

* **Cleaning:** removal of foreign material By water & soap.
  * precede Disinfection and sterilization (it's a pre-request for them)

* **Decontamination:** Reduction of microorganisms to a level at which items are safe to handle and can be used.
  * includes:
    - ↳ Cleaning → Decontamination By water and soap
    - ↳ Disinfection → Decontamination with spores remaining.
    - ↳ Sterilization → Decontamination with killing everything even the spores.

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* Pt.2 → continuing sterilization methods :)

* physical methods of Sterilization :

1. Moist Heat Above 100°C (Autoclave)

↳ Basic Mechanism: H_2O boild by heat → release steam → steam Condensed & then generate pressure on water → due to ↑ pressure water Temp will increase above 100°C → acheive Sterilization.

↳ Types of Autoclaves:

1. The Conventional autoclave (old one):

* Components: 1. Container that has water inside with electrical teild to heat water, Also, it contains air above.
2. Shelf to put the object we want to sterilize on it & it's porus to allow the steam to pass throug it.
3. pressure gauge: to measure pressure.
4. lid: the Cover 5. safety valve
6. Discharge tap: valve that releases the air.

* Working principle of this AutoClave: electrical power Heats up water & boil it → after boiling steam is released, inside the container there is air → the released steam pushes up the air from the container & get it out by the discharge tap (valve) till steam is remaining only in the container (replaces all the air), then valves close → steam remains & condenses then generates pressure on the water & as a result → water Temp increases above 100°C
  - ↳ Reaching 121°C At 2atm pressure within 20 min. if pressure got increased to 3 atm, Temp reach 134°C in 6 min (high temp in short time) but this is dangerous, so they get back to the 2atm & it's sufficient to acheive sterilization.

2. Prevacuum autoclave (new, developed)
3. Gravity displacement autoclave → uses steam from external source & replaces the air with it.

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> 1. Prevacuum Autoclave: This modern type of autoclave removes air from the chamber before steam is introduced, usually by creating a vacuum. By eliminating air first, the steam can reach all surfaces of the load quickly and effectively, leading to faster and more thorough sterilization.
>
> 2. Gravity Displacement Autoclave: In this type, steam enters the chamber from an external source and naturally rises to the top, gradually pushing the air downwards and out of the chamber through a vent. Steam is less dense than air at the same temperature, so it fills the chamber from the top down. This process effectively displaces the air with steam, but it takes a bit longer than the prevacuum method to achieve complete sterilization.
>
> The key to both methods is moist heat above 100°C, which denatures proteins in microorganisms, killing them and ensuring sterilization.

↳ mechanism of killing: 1. Denaturation 2. Coagulation

↳ used for sterilization of: 1. Surgical instruments & dressings.
2. Bed linen.
3. Cotton & gause.

↳ Advantages: 1. Latent heat so it has high penetration
2. Non-Toxic
3. Rapid (only 20 mins)

↳ disadvantages: 1. Not suitable for heat sensitive objects.
2. Sterilized objects remain moist
↳ specially surgical instruments → moist heat → water drops might remain on them Causing rust, unless it had drying technique it can be used otherwise, other methods more preferable for surgical instruments.

↳ Methods for monitoring of steam sterilizer (How to make sure it acheived sterilization correctly)

1. **Mechanical Indicators**: tells Temp, pressure, time so we know if sterilization acheived or Not.
2. **Chemical Indicators**: use filter paper with certain Chemicals on it (colorless before) → put it in autoclave, if color changed it means sterilization has been acheived.
3. **Biological Indicators**: use with the object a Spore forming bacteria then into autoclave, after it finished → take it & do subculture if the Bacteria grew, this means sterilization wasn't acheived, if didn't grow, this means sterilization has been acheived. eg of Bacteria can be used as Biological indicator: *Geobacillus Stearothermophilus

2. Dry Heat:

↳ Methods of Dry heat:

1. **Incenireation**: Burning of Contamination Materials.

2. **Direct Flame**: used to sterilize: loops (before Culturing) & points of Forceps.
↳ Should reach (Red Flame) ⇝ once it's reached this means sterilization was acheived.

3. **Hot air Oven**: acheive sterilization at 2 degrees: - At 160°C for 2 hrs.
- At 170°C for 1 hr

* used for sterilization of: 1. Glass-ware 2. Surgical instruments (this Method prefered over autoclave)
3. Oils 4. Powders

* Advantages: 1. Non-toxic.
2. Inexpensive.
3. Non-corrosive.

* Disadvantages: 1. Slow heat penetration
2. Time consuming.
3. Not suitable for heat sensitive objects.

3. Radiation: Emitted from Radioactive Cobalt 60 ⇝ Release (Gammarays)
↳ mechanism of killing: Breaks DNA.
↳ used in: 1. gloves 2. Catheters 3. surgical sutures

4. Filtration: Remove microorganisms from Biological fluids. eg. Plasma, Serum, harmones and Vitamins.
↳ these Filters has pores with diff sizes to retain microorganisms while allowing fluids to pass through.
* Filtration membranes usually made of: **Cellulose nitrate & polyster.**
↳ filtration membrans are layers used to filter out Particles & microorganisms: they have pores that allow fluid to pass through but trap larger particles or organism.
* there are virus filters as well that has very tiny pores to trap viruses as they're very small in size.
* HEPA (High efficiency particle arresters) used in: operation room & Drug filling cubicles.

✱ Chemical methods of Sterilization :

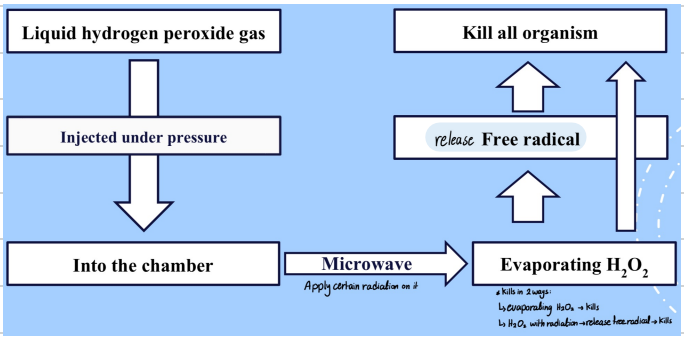
1. Gasuous Chemicals :

↳ plasma gas sterilizers : ✱ Plasma: (ionized gas) gas exposed to radiation & got ionized forming Cloud of gases with ions & electrons → Can act as sterilizer.

↳ types:

1. Hydrogen peroxide gas sterilizer

Mechanism →



2. Ethelene oxide gas sterilization

↳ highly lethal Alkylating agent

↳ Can make alkyl group to compounds
Alkyl = Alkane - H Atom

✱ item exposed to EO at 55°C for 3-6 hrs then it's aerated for 8-12 hrs to remove any trace of the gas (Disadvantage → takes long time)

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1. Hydrogen Peroxide Gas Plasma:
   - Mechanism: Hydrogen peroxide gas is converted into a plasma state by applying an electric field, which ionizes the gas, creating free radicals and reactive particles. These reactive particles can destroy microorganisms by disrupting cellular components such as DNA, proteins, and cell walls.
2. Ethylene Oxide Gas Sterilization:
   - Mechanism: Ethylene oxide gas sterilizes by alkylating the amino, carboxyl, and hydroxyl groups in proteins and DNA of microorganisms. This process prevents cells from reproducing and functioning, leading to cell death. Ethylene oxide is effective but requires aeration after sterilization, as it can be toxic to humans.
3. Peracetic Acid Gas Sterilization:
   - Mechanism: Peracetic acid is a strong oxidizer, and in its gaseous form, it reacts with essential cell components of microorganisms, including enzymes and structural proteins, which leads to cell death. It's effective for sterilizing sensitive equipment, especially in medical settings.

In summary, plasma gas sterilizers use reactive particles generated by ionizing gases, creating plasma. These reactive particles then kill microorganisms through chemical reactions with cellular components, ensuring effective sterilization.

3. Peracetic acid gas sterilization (Best Sterilant ☺)

↳ Peracetic Acid gas Composed of: Acetic Acid + hydrogen peroxide

↳ mechanism: 1. Denaturation 2. Oxidation (enzymes) 3. Disrupt Cell wall

↳ used for: 1. Heat sensitive devices, e.g: 1. Plastic 2. laparoscopes 3. Arthroscopes

↳ Advantages: 1. Non-Toxic. 2. suitable for heat sensitive objects.

↳ disadvantages: takes Much time → Total time of sterilization cycle is About 50 min.

2. Liquid Chemicals :

↳ types:

1. Peracetic acid (liquid) & Gluteraldehyde 2%: Considered High level disinfectant → if used for 20 mins.
   Considered Sterilant → if used for 10 hrs.

Good luck ☺