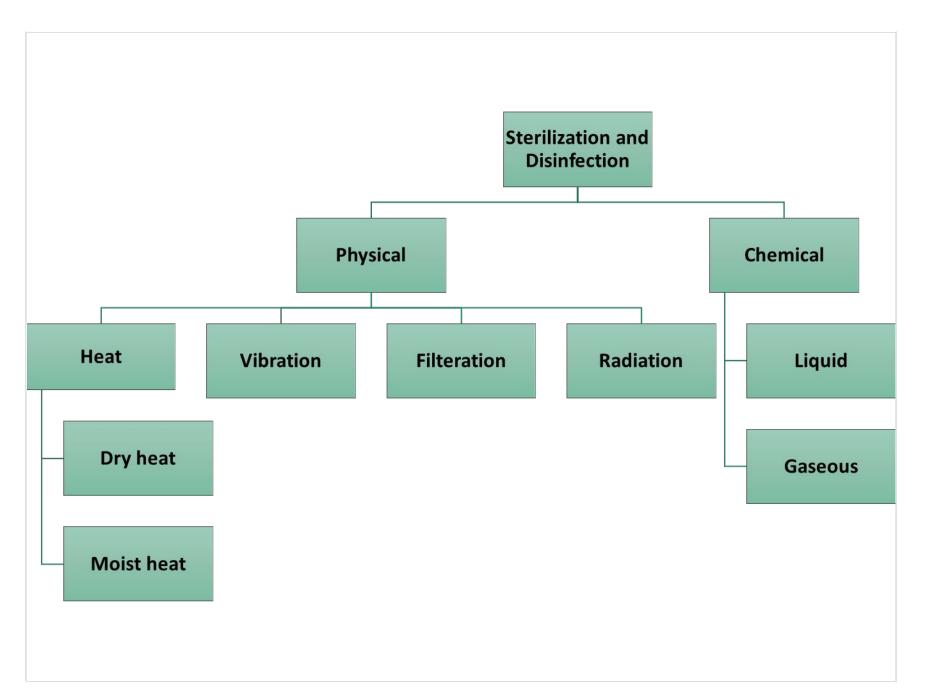
Sterilization & Disinfection

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Definitions

• Sterilization: complete killing of all forms of microorganisms, including bacterial spores

- Disinfection: killing or removing of harmful microorganisms. (Spores may not killed)
- Germs load ?



Methods of Sterilization

1. <u>Physical methods</u>:

- Heat (dry heat & moist heat)
- U.V. Light ... Sun light ?
- Irradiation:
- Filtration
- Sonic And Ultrasonic Vibrations.

PHYSICAL METHODS

I- HEAT : Most important method. Should be used whenever possible.

Action of heat: Cause biochemical changes in the cell's organic molecules such as protein denaturation, oxidative damage, and loss of water.

(More heat exposure time = more killing).

A-Dry heat at temperature of 160-200°C for 1-2 hours.
B- Moist heat eg. Pasteurization, Boiling, and
Autoclaving at 121 or 134 C for 10 or 15 minute.

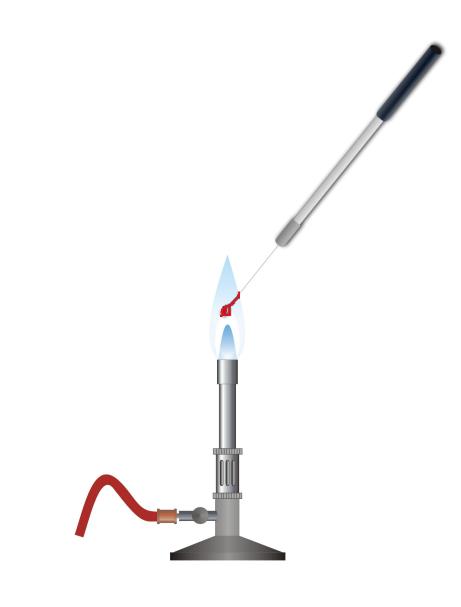
Moist heat: Faster and more efficient than dry heat because water molecules conduct heat better than air.

A- Dry Heat:

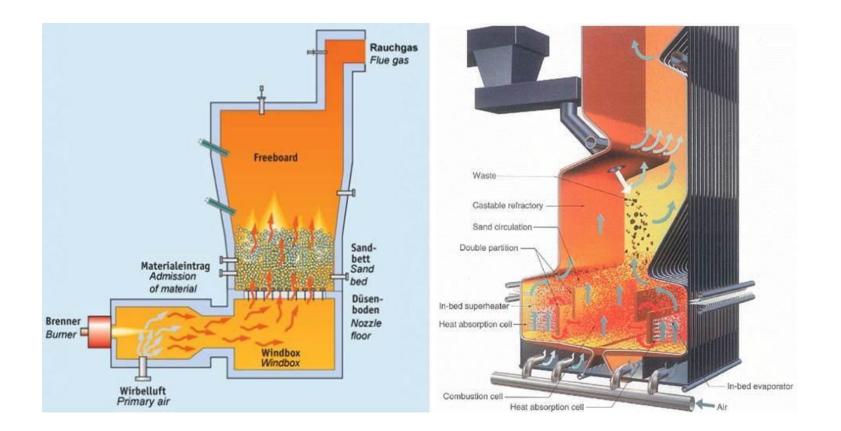
- •Flaming: used for sterilization of heat resistant objects such bacteriological loops and tips of forceps by passing them on Bunsen flame.
- •Incineration: This is a method of destroying contaminated material by burning them in an incinerator.
- •Hot air oven: Materials are exposed to high temperature (160 -200 C) for duration of one hour in an electrically heated oven.
 - Used for instruments (like forceps, scalpels, scissors), glasswares (such as petri-dishes, pipettes, flasks).
 - Hot Air does not penetrate materials easily and thus, long periods of exposure to high temperature are necessary.

• Oven





Incinerator



• <u>http://archive.sswm.info/category/implementation-tools/wastewater-treatment/hardwar</u> <u>e/solid-waste/incineration-large-scale</u> **B- Moist heat:** Faster and more efficient than dry heat because water molecules conduct heat better than air.

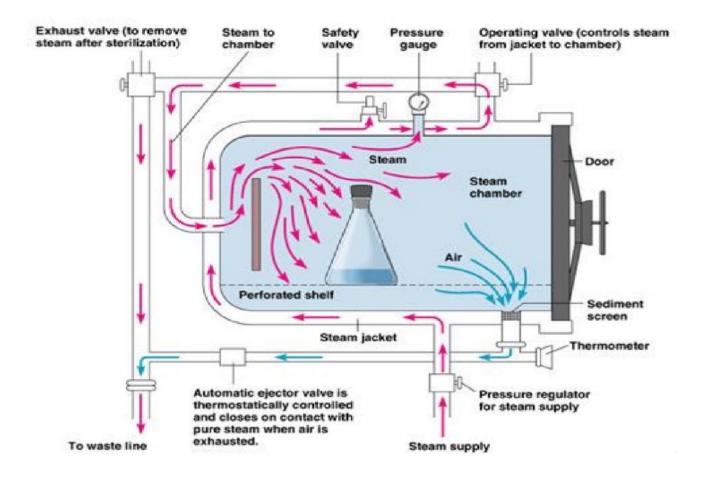
- •Boiling: Boiling water (100°C) kills most vegetative bacteria and viruses immediately. Some bacterial spores are resistant to boiling and survive.
- •Pasteurization: (by Louis Pasteur). There are two methods of pasteurization,
 - <u>conventional method</u> (heated at 63°C for 30 minutes)
 - <u>flash method</u> (heated at 72-74 °C for 15 seconds) followed by quickly cooling to 13°C.

Autoclave:

- □Water boils at 100°C at atmospheric pressure, but if pressure is elevated, water boiling temperature also increases.
- In the autoclave the water boils in a closed chamber. As the pressure rises, the boiling point of water also raises. At a pressure of 15 lbs inside the autoclave, the temperature is 121°C. Exposure of materials and media to this temperature and steam for 15 minutes sterilize them including spores.
- This method is very efficient compared to other methods.

Autoclave:





II- Irradiation: By exposing organisms to ionizing or non-ionizing radiation types.

- Non-ionizing rays has low energy and is poor in penetration power (UV radiation, Infrared radiation).
- Ionizing rays are high-energy rays with good penetration power (X rays, Gamma rays).

Can sun light kill Bacteria and how ?

III- Filtration: it works by separating microbes without killing them. Membrane filters with pore sizes between 0.2-0.45 μ m are usually used to separate microbes from solutions that can not be autoclaved.

Viruses can pass through ?

IV- Sonic And Ultrasonic Vibrations: Sound waves of at very high frequencies >20,000 cycle/second kills bacteria and some viruses on exposing for one hour.

The killing effect is due to the friction and heat generated during sonication. This method is not reliable since many viruses and phages are not affected by these waves.

2. Chemical methods:

(used for heat sensitive materials).

- Disinfection: killing or removing of harmful microorganisms. (Spores may not killed)
- **Disinfectants**: chemicals that can achieve disinfection. They can destroy pathogenic bacteria from inanimate surfaces.
 - narrow spectrum of activity
 - very wide spectrum of activity.
- **Antiseptics** Chemicals that can be safely applied over skin and mucus membranes and kill pathogenic organisms. They prevent the growth of microorganisms either by destroying them or by inhibiting their growth and metabolism. Example 70% ethanol
- An antiseptic is applied to the body, while disinfectants are applied to nonliving surfaces,
- A 70% aqueous solution of ethanol is more effective at killing microbes than absolute alcohols. Why?.

Common disinfectants include alcohols, quarternary ammonium salts, formaldehyde and glutaraldehyde, bleach.

Some antiseptics are: hydrogen peroxide, iodine, polyhexanide, and povidone-iodine, alcohol, boric acid, and benzalkonium chloride.

Classification of disinfectants:

- Based on consistency: a. Liquid (E.g., Alcohols, Phenols), b. Gaseous (Formaldehyde vapor).
- Based on spectrum of activity: a. High level , b. Intermediate level, c. Low level
- Based on mechanism of action:
 - a. Action on membrane (E.g., Alcohol, detergent),
 - b. Denaturation of cellular proteins (E.g., Alcohol, Phenol),

c. Oxidation of essential sulphydryl groups of enzymes (E.g., H2O2, Halogens),

d. Alkylation of amino / carboxyl/ and hydroxyl group (E.g., Ethylene Oxide, Formaldehyde),

e. Damage to nucleic acids (Ethylene Oxide, Formaldehyde)

- Q- How do we know that a disinfection or sterilization process is efficient ?
- Q- Why autoclaving is very good method for sterilization compared to other methods?
- Can Sun Kill microorganisms?
- Why moist heat is more efficient than dry heat in sterilization?
- Does filtration method, using 0.45 um filters, removes viruses from a solution ?
- How do you sterilize plastics ?
- How do you disinfect lab bench ?



