

STERILIZATION AND DISINFECTION



Importance of hand washing shown by Semmelweis

STERILIZATION

- A physical or chemical process that destroys or eliminates all forms of microbial life, including spores.
- A satisfactory sterilizing process may be regarded as one that achieves a high probability of sterility i.e. a process that kills more than 10^6 organisms ,including spores of defined exceptionally high degree of resistance.

METHODS OF STERILIZATION

- Determined by the character of items to be sterilized.
- Choice of process requires knowledge of the equipment and of stability of the material to be sterilized.

CLASSIFICATION

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graph TD; A[CLASSIFICATION] --- B[ ]; B --- C[PHYSICAL]; B --- D[CHEMICAL]; C --- E["1) DRY HEAT<br/>2) STEAM STERILIZATION<br/>3) INCINERATION<br/>4) FILTERATION<br/>5) RADIATION"]; D --- F["1) ETHYLENE OXIDE<br/>2) GLUTARALDEHYDE"];
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PHYSICAL

- 1) DRY HEAT
- 2) STEAM STERILIZATION
- 3) INCINERATION
- 4) FILTERATION
- 5) RADIATION

CHEMICAL

- 1) ETHYLENE OXIDE
- 2) GLUTARALDEHYDE

PHYSICAL METHODS

DRY HEAT STERILIZATION

- For heat stable materials
- Sterilization effect is due to enzyme inactivation, protein denaturation or both.

- **Red heat:** Inoculating wires and loops, points of forceps ,surface of searing spatulas.
- **Hot air sterilizers:** Glassware, glass syringes, glass pipettes.
- British standards(BS3421)
- Should be fitted with a fan that provides forced air circulation throughout the oven chamber, a temperature indicator, a control thermostat and timer, open mesh shelving and adequate wall insulation.

STEAM STERILIZATION

- **AUTOCLAVE:** Provides moist heat at temperatures above 100°C by exposing the load to saturated steam at pressures greater than atmospheric.
- As steam condenses on the cooler load, it releases both thermal energy and moisture ,which together denature all microbial proteins.



AUTOCLAVE

TIME AND TEMPERATURE REQUIRED FOR STERILIZATION IN THE AUTOCLAVE

Sterilizing temp (°C)	Pressure above atmospheric (bar)	Sterilization hold time (min)
121-124	1.1	15
134-138	2.2	3

TYPES OF AUTOCLAVES

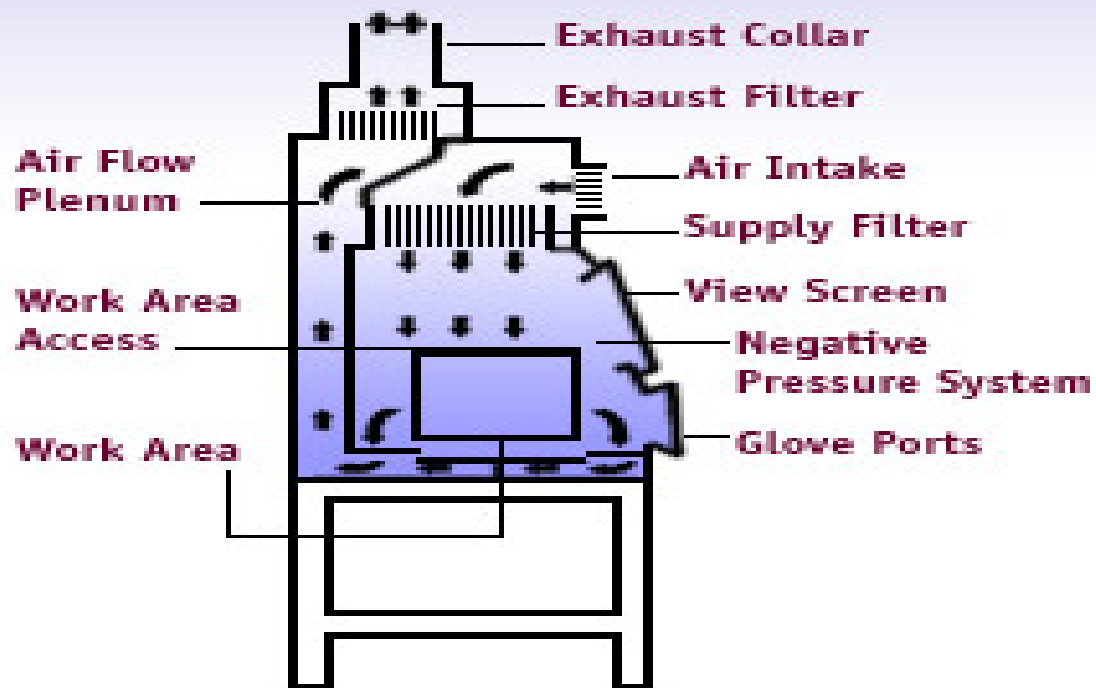
- 1) Simple laboratory autoclave
- 2) Transportable bench top autoclaves
- 3) Large simple autoclaves
- 4) Downward displacement autoclaves
- 5) Multipurpose laboratory autoclaves.

STERILIZATION BY FILTRATION

- 1) Membrane filters
- 2) Syringe filters
- 3) Vacuum & “in line” filters
- 4) Pressure filtration
- 5) Air filters- HEPA



Total Exhaust



STERILIZATION BY RADIATION

- IONIZING radiation - γ radiation from radioactive elements , usually Co^{60}
- ULTRAVIOLET RAYS: Mercury vapour lamps emitting radiation in the range of 250-260nm are bactericidal & to a lesser extent sporicidal.



CHEMICAL METHODS

1) ETHYLENE OXIDE: Medical & surgical articles.

Highly lethal gas.

Attaches to sulphhydryl bonds & proteins thereby interfering with their structure and function.

Effect most rapid at 30-40% relative humidity.

DISADVANTAGES: Toxic and highly explosive.

CHEMICAL METHODS (CONTD.)

2) **Glutaraldehyde:**

- Chemically related to formaldehyde but is 2-8 times more sporicidal.
- In a 2% alkaline aqueous solution it is sporicidal at room temperature
- Used to sterilize plastics, rubber & delicate lensed instruments

DISADVANTAGES

- Significant loss of activity in the alkaline state (over a 2 week period).
- Exposure of at least 10 hours required.
- Need to remove residual glutaraldehyde by rinsing with sterile water.

3) **Gas plasma:**

A very small quantity of hydrogen peroxide in various phases, including a low temperature gas plasma excited by radio waves .Sporicidal, bactericidal ,virucidal.

BIOLOGICAL CONTROLS OF DIFFERENT STERILIZATION METHODS

METHOD OF STERILIZATION	BIOLOGICAL CONTROL
Hot air oven	<i>Bacillus subtilis</i> subsp. <i>niger</i>
Autoclave	<i>Bacillus stearothermophilus</i>
Ethylene oxide	<i>Bacillus globigi</i>
Ionizing radiations	<i>Bacillus pumilis</i>
Filtration	<i>Serratia marcescens</i> , <i>Pseudomonas diminuta</i>

CHEMICAL CONTROL

- A Browne's tube containing a red solution is placed within the load. A change of colour of the solution to green indicates proper sterilization.

DISINFECTION

- It is a process whereby pathogenic organisms, but not necessarily all microorganisms or spores are destroyed. Disinfectants are used in hospitals and clinical laboratories for three main purposes :
 - 1) To render contaminated objects safe for further use.
 - 2) To reduce the microbial contamination of the inanimate environment.
 - 3) To prevent spread of microorganisms by contaminated wastes.

COMMON DISINFECTION PROCEDURES

- PHYSICAL

- 1) Hot water or steam

- a) At temperature below 100°C -
Pasteurisation

- b) At temperature of 100°C –Boiling/Free steam/Tyndallization

- c) At temperature above 100°C -
Autoclaving

- 2) Ultraviolet rays

- 3) Mechanical means Filtration

CHEMICAL

- 1) Phenols & cresols
- 2) Halogens
- 3) Metallic salts
- 4) Aldehydes
- 5) Alcohols
- 6) Dyes
- 7) Vapour phase disinfectants.
- 8) Surface active disinfectants.

COMMON DISINFECTANTS AND THEIR CONCENTRATIONS

DISINFECTANT	CONCENTRATION
Ethanol	70%
Methylated spirit	70%
Glutaraldehyde	2%activated(Cidex)
Bleaching powder	14g/l of water
Sod.hypochlorite	1%,0.1% solution
H ₂ O ₂	3%solution
Lysol	2.5% solution
Savlon	2.0%,5%

TESTING OF DISINFECTANTS

1) Minimum inhibitory concentration (MIC)

2) Phenol coefficient

- Rideal Walker

- Chick Martin

3) Capacity test (Kelsey & Skyes test)

ANTISEPSIS

- It is the use of a substance that prevents or arrests the growth or action of microorganisms either by inhibiting their activity or by destroying them.
- The term “antiseptic” is used especially for preparations applied to living tissue.

AREAS OF USE

- AIM OF ANTISEPSIS- Reduction or destruction of undesirable microorganisms on skin & mucosal surfaces while preserving the normal, resident flora.

REQUIREMENTS ON ANTISEPSIS & ANTISEPTICS

- 1) Lack of skin/mucosal irritancy
- 2) Lack of systemic toxicity (usually measured as oral toxicity)
- 3) Lack of teratogenic, mutagenic & carcinogenic effects)
- 4) Absorbed blood levels in man must be far below the toxic levels for both long & short term exposure.
- 5) Stability in both concentrated & use –dilution forms.

COMMONLY USED ANTISEPTICS

- 1) Heavy metals: Mercurials, silver nitrate, silver sulfadiazine
- 2) Bisphenols: Hexachlorophene
- 3) Alcohols: Ethanol at concentration of 70-90%
- 4) Iodine & iodophors
- 5) Quaternary ammonium compounds
- 7) Phenols

All the best..