Basic Bioscience

Control of Microbes

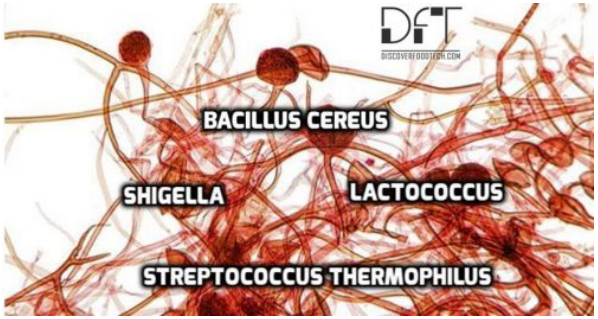
Dr Azham Zulkharnain

Microbial Growth Control - Introduction

- Microbes present a constant threat to health, jeopardize our food supplies and destroy many useful materials.

- Fortunately, microbes can be eliminated or reduced in number through the use physical or chemical methods.

- These protective measures are particularly important in the hospital, where high concentrations of potentially pathogenic organisms are found.



Nosocomial infections

Types of microbial growth control:

1. Inhibition:
   Microbes or their undesired activities are inhibited by bacteriostatic compounds or techniques.

2. Sterilization:
   Microbes themselves are killed by bacteriocidal compounds or techniques.

Considerations involved when dealing with microbial control:

1. Areas of treatment or application
2. Types of control/treatment to be utilized

Food Control

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Effect of detergents as antibacterial agents on biofilm of antibiotics-resistant *Vibrio parahaemolyticus* isolates

N. Elexson[a], L. Afsah-Hejri[a], Y. Rukayadi[a], P. Soopna[a], H.Y. Lee[a], T.C. Tuan Zainazor[c], M. Nor Ainy[a], Y. Nakaguchi[b], N. Mitsuaki[b], R. Son[a]

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http://doi.org/10.1016/j.foodcont.2013.07.020

Efficiency of Detergents against Microbial Biofilm Growth in Kuching, Sarawak

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Biofilm of Antibiotics Resistant Salmonella Typhimurium and Salmonella Enteritidis Against Detergents

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Publication date	2016
Description	1 Department of Molecular Biology, Faculty of Resource Science and Technology, University Malaysia Sarawak, 94300, Kota Samarahan, Sarawak, MALAYSIA 2 Food Safety Research Center (FOSREC), Faculty of Food Science and Technology, Universiti Putra Malaysia, 43400 UPM Serdang, Selangor Darul Ehsan, MALAYSIA *Corresponding author. E-Mail: nelexson@frst.unimas.my; Tel: +06082532979; Fax: +06082583160 ... Received: 4 March 2016 Revised: 31 May 2016 Accepted: 6 June 2016 Online: 1 August 2016
Scholar articles	Biofilm of Antibiotics Resistant Salmonella Typhimurium and Salmonella Enteritidis Against Detergents E Nillian, Y Rukayadi, S Radu - 2016 Related articles

Microbial Growth Control - Introduction

1.Areas of treatment or application.

- Treatment of inanimate (tabletop/working surfaces) objects.

1.Decontamination removes or kills most microbes, making an object or surface safe.

2.Disinfection removes most or all pathogens from an object or surface.

3. Sterilization removes or kills all microbes.

- Treatment of living organisms or tissues

1.Agents used to treat living organisms or tissues must be nontoxic as well as antimicrobial.

2.Antiseptic compounds can be applied to tissues.

3.Antimicrobial agents can be taken internally.

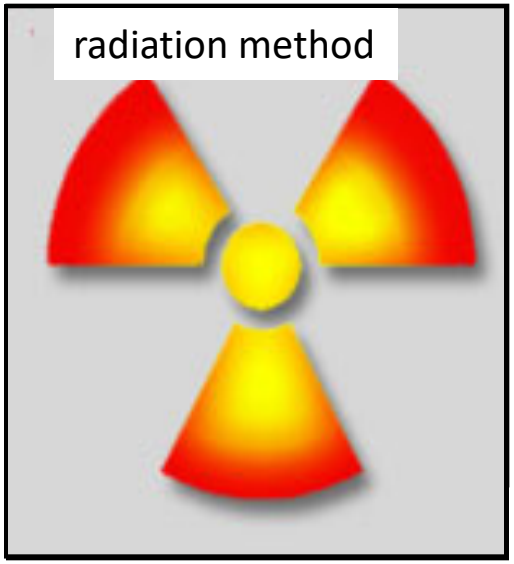
2.Types of control/treatment to be utilized

- Basically, there are three principles involved in the control of microbial control:

1. Physical methods.
2. Chemical methods.
3. Antibiotics.

Microbial Growth Control - Physical Methods

- Physical methods of microbial growth control can be divided into four types :
  - Heat – most widely used
  - Filtration
  - Radiation
  - Ultrasonic vibrations



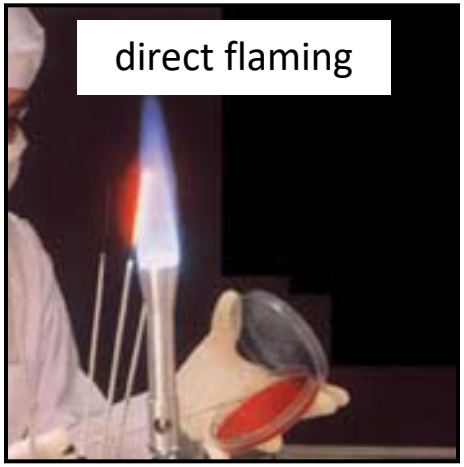
Heat is mostly used because :

- Useful for inanimate objects, effective and least expensive.
- Lethal effects of heat on microbes may be due to:
  - Protein denaturation (coagulates) and enzymes.
  - Effects on membrane fluidity
  - Denaturation of other macromolecules
- Two types of heat sterilization:
  - dry heat methods
  - moist heat methods

Physical Methods - Dry Heat Methods

a. Direct flaming

- Example: inoculation loop.


direct flaming

b. Incineration

- Burning to ashes/oxidation.
- Must be complete.
- Used to destroy disposable items, soiled dressings, tissue specimens.
- Example: Hospital disposables and specimens.


laboratory oven for dry heat sterilization

c. Hot-air sterilization

- Baking in oven.
- Very effective method of sterilization, but requires temperature of 170 ^{o}C for ~ 2 - 3 hr.
- causes oxidation of microbes, sterilizes.
- used when moisture is undesirable - glassware, metals, powders or petroleum-based products (oils).

Physical Methods - Moist Heat Methods

a. Boiling

- 98 -100°C for 10 min.
- Inactivates most vegetative cells, not heat-resistant forms (i.e. endospores, some viruses & bacterial toxins).
- e.g. drinking water, canning jars, etc.

b. Tyndallization

- A sterilization process used for foods, especially in home canning.
- Foods are sealed and heated to boiling (this kills microbial cells, but generally not endospores).
- Food is then cooled and stored for 1 day at room temperature (endospores germinate to vegetative cells).
- The food is reheated (100°C) to kill those cells

Physical Methods - Moist Heat Methods

c. Pasteurization

- Heat material, holding at specific temperature for specific length of time, cool rapidly.
- Low temperature long term (holding) method: 62°C for 30 min.
- High temperature short term method: 72°C for 15 sec.
- Inactivates pathogens, reduces total microbial population, but does not sterilize.
- i.e. Dairy products, wine, beer, etc.

d. Autoclave

- Autoclaves uses steam under pressure (increases the steam temperature) for moist sterilization.
- Destroys all lifeforms – sterilization.
- Coagulate proteins and causes hydrolysis.
- The chamber contains 100% steam during sterilization process.

Physical Methods - Moist Heat Methods

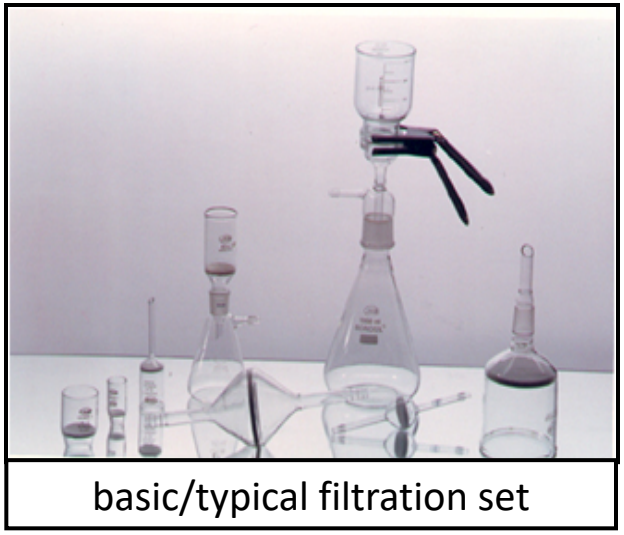
d. Autoclave

- During autoclave, the correct parameters are :
  - Pressure : 15 - 20 psi
  - Temperature : 115 - 121°C
  - Time duration : 15 - 20 min.
- Application : microbiological media, surgical instruments, etc.
- Autoclave cycle :
  - After the door is sealed and sterilization begins, the autoclave pressurizes and the temperature of the sample rises.
  - Large samples warm more slowly than small ones (large autoclave system may require hours for heating).
  - Aqueous samples boil during depressurization to cool (heat of vaporization) to 100°C (cooling must be slow to avoid boiling over).

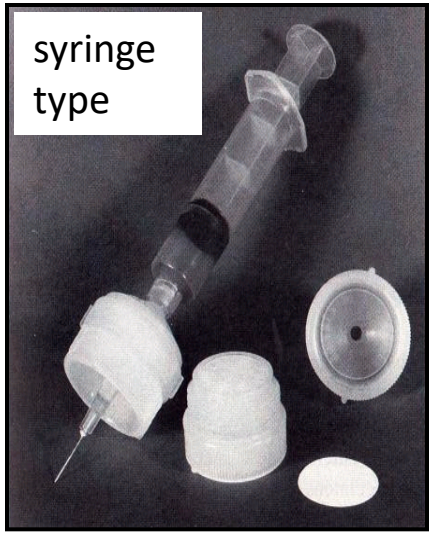
Physical Methods - Filtration

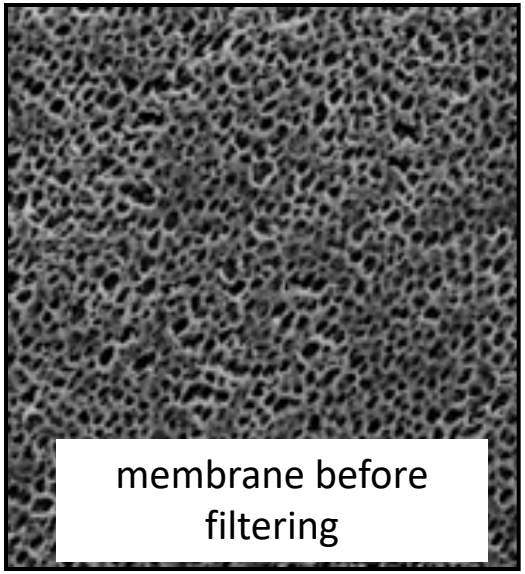
- Liquids can be sterilized by filtering out particles.

- Solids physically separated from liquids by passage through filters with extremely small pores (i.e. porcelain, ground glass, diatomaceous earth, asbestos, sand, membrane filters).

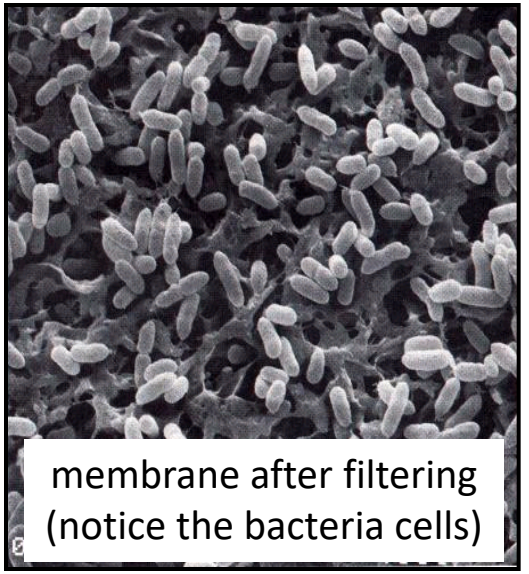


basic/typical filtration set





membrane before filtering



membrane after filtering (notice the bacteria cells)

Physical Methods - Filtration

- Membrane filters, made of microscopic and modified fibers, has small and consistently sized pores are normally used.
  - Filtration methods are used widely for :
- Heat-sensitive materials (media, medications) that can't be heated.
- Beer and wine.
- Swimming pools and spas.
- Sewage, air, testing water or air for organisms or allergens.

Disadvantages of filtration method :

- Not suitable for liquids such as milk (not a solution).
- Limitations in size of organisms to be removed (sand filters in water treatment remove only protozoal cysts and helminth eggs).
- Even nucleopore filters cannot remove viruses.

Physical Methods - Radiation

- Radiation cause lethal changes in DNA, denatures proteins (produces hyperactive ions and free radicals).
- Two types of radiation treatments :

  a. Non-ionizing rays (UV light)

    - UV radiation is often used to sterilize surfaces.
    - Causes dna damage including breakage of the dna backbone by the formation of thymine dimers.
    - Apparent limit - cannot penetrate materials (cloth, glass, paper, etc).

UV radiation is used to sterilize laminar flows

b. Ionizing rays (X-rays, gamma rays).

- Ionizing radiation is more effective and can penetrate materials.
- Ions (loss of electrons) are formed when they pass through molecules.
- Causes dna damage and production of toxic reactant products.
- Used to sterilize plastics, medications, foods (retards spoilage).

Physical Methods – Ultrasonic Vibration

- Ultrasonic vibrations uses high frequency sound waves from a machine called a sonicator.
- Killing microbes by shock waves that disintegrates cell wall and membranes.

Different models of sonicators

Physical Methods – Preservation Methods

a. Increased osmotic pressure.

- High concentrations of salt, sugar.
- Dehydrates cells, more effective against bacteria than fungi.
- Application : food preservation.

b. Desiccation

- Drying, removal of water.
- Retards growth, but does not always kill.
- More effective against bacteria than fungi.
- Application : food preservation.

Physical Methods – Preservation Methods

c. Acid, Alkaline pH

- Inactivates enzymes, inhibits growth.
- Does not always destroy microbes.
- Uses: acid pH (benzoic, sorbic, propionic acids) - food preservation.

d. Decreased temperatures

- Refrigeration.
  - 5 - 10^{o}C.
  - retards growth, does not prevent growth.
- Freezing
  - below 0^{o}C (-10^{o}C).
  - prevents growth, does not kill all organisms.