



Microbiology – Lecture 3 Sterilization



TEAM 437

Red: important

Green: doctor notes

Black: original slides

Grey: extra information

In this link, you will find any corrections or notes unmentioned in the team's work. Please check the link below <u>frequently.</u>

https://docs.google.com/presentation/d/1yIQt3G8UDFG6xYM RhXkTk-dS54NeTfhJaPe_y0M-kjk/edit?usp=sharing

Objectives



- 1- Define the terms sterilization, disinfectant and antiseptic.
- 2- Classify the different methods of sterilization (physical and chemical methods).
- 3- Know and realizes that heat is the most important method of sterilization and its application in medical practice.
- 4- Know dry heat as applied in hot air Oven and moist heat as applied in autoclaves.
- 5- Know the principles of Autoclave function and monitoring methods of sterilization.
- 6- Know the importance of non heat sterilization methods and their use for sterilization of heat sensitive objects.
- 7-Know the difference between antiseptics and disinfectants.
- 8- Know types and scope of function of the disinfectants and antiseptics and factors affecting their functions.
- 9- Know the medical applications of different disinfections and antiseptics.

Definition and sterilization methods

Physical methods

for heat sensitive

equipment.

Chemical method; used



Term	Definition
Sterilization	complete killing of all forms of microorganisms, including bacterial spores
Disinfection	killing or removing of harmful vegetative microorganisms.
Disinfectant	chemical substance used to achieve disinfection used on inanimate objects. Toxic to humans.
Antiseptic	disinfectant that can be safely used on living tissues

Methods of Sterilization
 Heat (dry heat & moist heat), most important and should be used whenever possible.
 U.V. Light, (ultra violet)

Ionizing Radiation

Ethylene Oxide

Gluteraldehyde

Filtration

Using very strong chemicals

Monitor k		Heat	Dry Metals, Glassware, Ointment / Oils/ Waxes/Powder	160°C /60 mints
			Moist heat	121° C /15 mints or 134° C /10 mints
y physi			Pasteurization For milk	74°C for 3-5 seconds or 62°C /30 mints
cal, o	A-Ph	UV Light	TB lab	254 nm waves
chemic	A-Physical	Ionizing Radiation	sterilization of disposable	X- rays, gamma rays & cosmic rays
A-Physical Chemical (heat sensitive) Monitor by physical, chemical or biological)	Ch ser	Ethylene Oxide (gas)	endoscopes	55-60°C /4-6 hours.
	B- emical (heat nsitive)	Gluteraldehyde (Aldehydes)	Endoscopes anesthetic tubes	2% 20 mints –TB 2 hrs
tion cal)		Filtration	For sera and antibiotics	0.22-0.24 μm pores
Disii		Phenolics fluids	Surfaces Surgical and neonal units	1-2%
nfect		Gluteraldehyde	surfaces	2%
2- Disinfectant		Alcohole	Surfaces/thermometer	
		Chlorhexidine	skin	0.5%
3- Antiseptic		lodine (halogens)	skin	10%
3- ptic		Alcohole	skin	70%

Level	uses	application	example
High-level disinfectants Kill all including spores	Used for items involved in invasive procedures but NOT withstand sterilization (critical)	Endoscopes, Surgical instruments	Moist heat Gluteraldehyde Hydrogen peroxide Chlorine dioxide Formaldehyde Peracetic acid
Intermediate-level disinfectants Kill all including mycobacterium non-enveloped viruses fungus and bacteria	Used for cleaning surface or instruments without bacterial spores and highly resilient organism (semicritical)	Laryngoscopes, Anesthesia breathing circuitsetc	Phenol compounds Alcohol Iodophor
Low-level disinfectants Enveloped viruses and bacteria	Used to treat noncritical instruments and devices, not penetrating into mucosa surfaces or sterile tissues (noncritical)	Hospital surfaces	Quaternary ammonium compounds

PHYSICAL METHODS COMMON HEAT METHODS



1.Dry heat 2.Moist heat

Dry Heat- kills microorganisms by destroying their oxidative processes. At temperature of 160C for 1 hour.

<u>Simplest method</u> is exposing item to be sterilized to the naked flame e.g. Bunsen burner- for sterilizing bacteriological loops, knives, blades. **Hot air oven expose items to 160 °C for 1 hour.**

Has electric element in chamber as source of heat plus a fan to circulate air for even distribution of heat in chamber. Oven without fan is dangerous. Used for items that are lacking water such as:

- a) Metals
- b) Glassware
- c) Ointment / Oils/ Waxes /Powder

Uses <u>hot water</u>. Moist heat kills microorganisms by denaturation proteins.

Autoclaving: The autoclave is a tough double walled chamber in which air is replaced by pure saturated steam under pressure.

standard sterilization method in hospitals.

The equipment is called Autoclave and it works under the same principle as the pressure cooker where water boils at increased atmosphere pressure i.e. because of increase pressure the boiling point of water is >100 °C.

Autoclaving and how it works

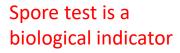


1. Physical method: use of thermocouple to measure accurately the temperature.

Monitoring of Autoclaves

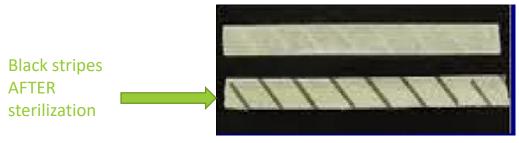
2. **Chemical method:** it consists of heat sensitive chemical that changes color at the right temperature and exposure time. e.g:

- a)- Autoclave tape
- b)- Browne's tube.



3. **Biological method**: where a **spore**-bearing organism is added during the sterilization process and then cultured later to ensure that it has been killed.

This is autoclave tape; it will have NO black stripes before sterilization.







Advantages of Autoclave

Temp. > 100 C therefore spores killed.

Condensation of steam generates extra heat.

The condensation also allows the steam to penetrate rapidly into porous materials.

Note: for all invasive procedures at operating room or clinics, autoclavable equipments should be used.

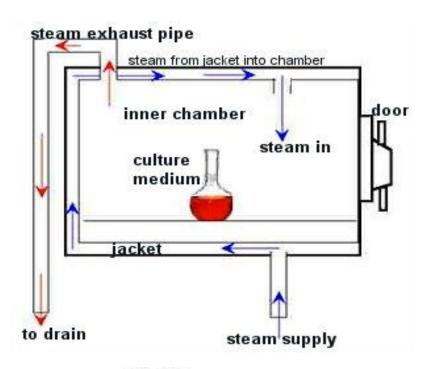
How does it work?

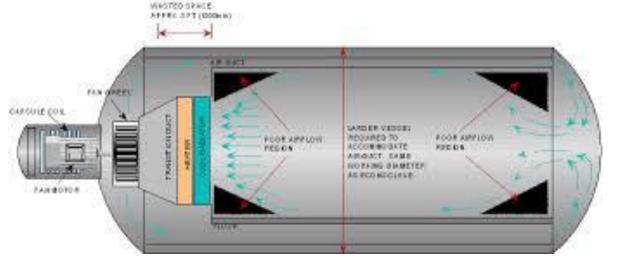
The air in the chamber is evacuated and filled with saturated steam. The chamber is closed tightly the steam keeps on filling into it and the pressure gradually increases. The items to be sterilized get completely surrounded by saturated steam (moist heat) which on contact with the surface of material to be sterilized condenses to release its latent heat of condensation which adds to already raised temperature of steam so that eventually all the microorganisms in what ever form are killed.

The usual temperature achieved is $121 \,^{\circ}\,$ C, exposure time of only 15 minutes (or 134 C for 10 minutes).









In short pasteurization:
Is the most
effective way to
sterilize.
-it requires high
temperature and
high pressure.
-it does the work
at short time.

Moist heat: Other Applications



A) Pasteurization

Used heat at temperatures sufficient to inactivate harmful organism in <u>milk</u>. The temperatures of sterilization is <u>not achieved</u>.

Temperature may be

74°C, for 3-5 seconds. (Flash methods)

62°C for 30 minutes. (Conventional method).

Why do we pasteurize foods?

To prevent transmission of zoonotic diseases to humans, like:

- Typhoid fever
- Brucellosis
- Tuberculosis
- Q fever

B) Boiling

quite common especially in domestic circumstances.

C) Steam Sterilizers

For babies' bottles.

Other Physical Methods: Radiation



UV Light

 Has limited sterilizing power because of poor penetration into most materials. Generally used in irradiation of air in certain areas such as operating rooms and tuberculosis labs.

Ionizing Radiation

• e.g. *Gamma radiation*:
has greater energy than
U.V. light, therefore more
effective. Used mainly in
industrial facilities e.g.
sterilization of disposable
plastic syringes, gloves,
specimens containers and
Petri dishes.









- Use of membrane filter. Example; membrane filter made of cellulose acetate.
 Generally removes most bacteria but viruses and some small bacteria e.g.
 Chlamydia & Mycoplasma may pass through (require smaller pore size).
- Thus filtration does not technically sterilize items but is adequate for circumstances under which is used.
- Main use: for heat labile substances e.g. sera, antibiotics. Other use: AC of certain operating theatres.



Chemical Methods

Some strong chemical substances

may be used to achieve sterilization (kill spores) e.g.

Gluteraldehyde and Ethylene

oxide. Used for heat sensitive

equipment and materials e.g.

lensed endoscopes and plastics.



Ethylene Oxide Chamber

Ethylene oxide alkylates DNA molecules and thereby inactivates microorganisms.

Temperature: 55-60° C and exposure period 4-6 hours.

Activated alkaline Gluteraldehyde 2%

Immerse item in solution for about 20 minutes.

If *Mycobacterium tuberculosis* or spores present then immersion period is 2-3 hours.

Chemical Methods

Other uses: Hypochlorite (at certain concentrations) used for drinking water supply, house cleaning and swimming pools.

Note; Clorox is a tamer type of hypochlorite

Disinfectants / Antiseptics

eg. phenolics, chlorhexidine, alcohol, iodine, etc..

Factors Influencing Activity of Disinfectants



- 1. Activity directly proportional to temperature.
- 2. Directly proportional to **concentration** up to an optimum concentration. After this level no advantage in further increases in concentration.
 - 3. Disinfectants may be inactivated by:
 - A) Dirt
 - B) Organic matter: Proteins, Pus, Blood, Mucus and Feces.
 - C) Non organic: Cork, Hard water and Some plastics.
- 4. Time: Disinfectants need time to work.
- 5. Range of Action: Disinfectants not equally effective against the whole spectrum of microbes. e.g. Chlorhexidine less active against Gram negative bacteria than Gram positive cocci. Hypochlorite and Gluteraldehyde are more active against hepatitis viruses than most other disinfectants.





Note; it must be clean before disinfecting

Summary: Disinfectants & Antiseptics



Disinfectants/	GPC	Activity against			Inactivated by		Corrosive
Antiseptics		GNB	Spores	TB	Protein	Soap	Action
Phenolics Sudol	++	++	-	+	±	-	+
Izal	++	++	-	ı	<u>±</u>	_	_
Soluble Phenolic* e.g. clearsol	++	++	-	+	±	-	<u>+</u> to +
Chlorine compound	++	++	++	+	++	-	_(buffered Φ++ or <u>+</u> Solution)
Lodophor	++	++	+ (Slow)	+	+	-	-
Chlorhexidine (Hibitane)	++	+	-	1	-	-	-
70° alcohol	++	++	=	±	++	_	-
Formaldehyde	++	++	++ (<u>slow)</u>	++	+	-	++
Gluteraldehyde (Cidex)	++	++	++	++	±	-	+





Floors, walls

Phenolics fluids 1-2%

Surfaces tables

Hypochlorite, Alcohol

Skin:

Surgeons' hands

• Chlorhexidine, Iodine alcohol

Patient's skin

• 70% Alcohol, Iodine

Endoscopes

• Gluteraldehyde 2% (Cidex), sub-atmospheric steam

Thermometers

• 70% Alcohol



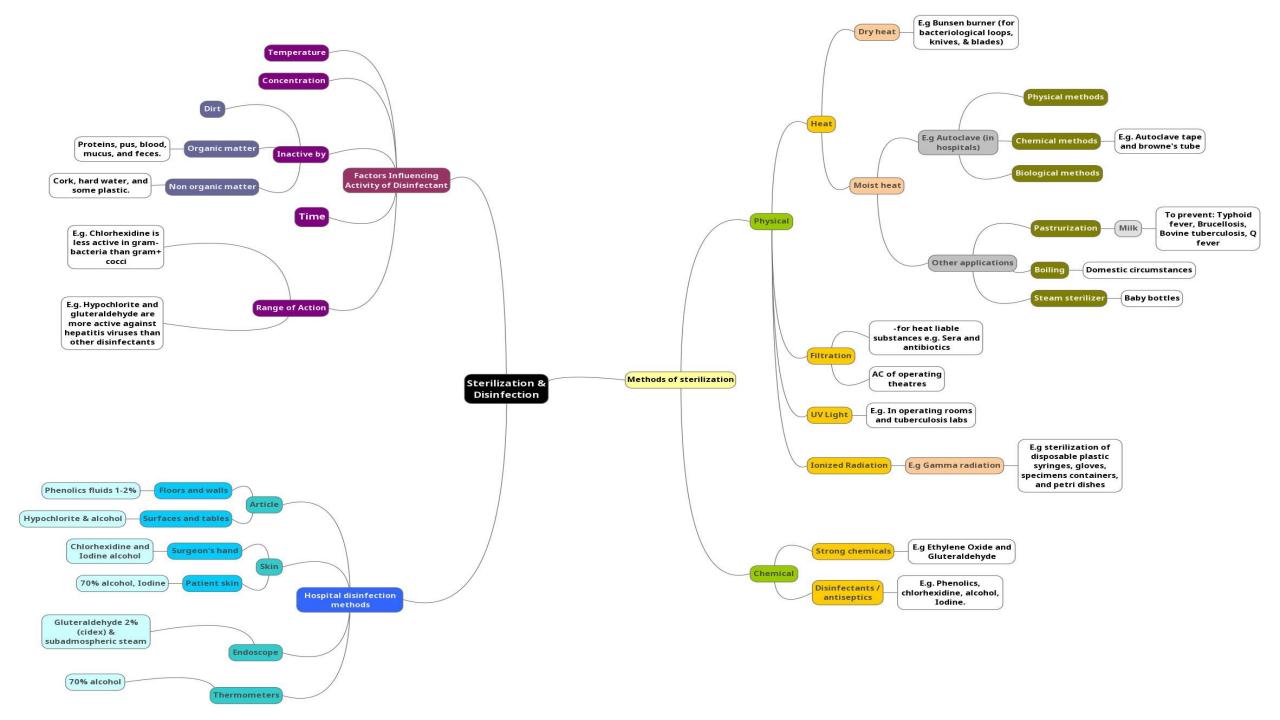
Important Points

- Any instrument or item used for sterile body site should be sterile.
- Any instrument or item used for non-sterile body site can be disinfected.

Soap, Water and common sense are yet the best antiseptics

William Osler







b-conventional method

6- 74C, for 3-5 seconds is called:

A-frozen method



1- killing or removing of h	narmful vegetative microorg	anisms, is:				
A- sterilization	b- disinfectant	c- antiseptic	d-disinfection			
2- Disinfectants may be inactivated by :						
A- water	b-dirt	c- soap	d- washing			
3-Gamma radiation: has greater energy than U.V. light, therefore more effective. Used mainly in :						
A-industrial facilities	b-tuberculosis labs	c-Browne's tube	d-Patient's skin	Answers		
				D		
4-We use pasteurization t	to prevent the transmission	of which disease?		В		
A-tuberculosis	b-small pox	c- hepatitis	d-liver failure	А		
				А		
5-Chlorhexidine less activ	e against Gram:			В		
A-positive	b- negative			С		

c-flash method

Sherries Medical Microbiology, an Introduction to Infectious Diseases.

d-pasturized method



MICROBIOLOGY 437

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