Practical Principles of Microbial Physiology // 4th Stage **Lab 6**
Effect of Ultraviolet radiation on microbial growth **3 pages**

Radiation

- **Radiation** is a form of energy that travels in rhythmic waves through space or a material medium, the distance between the crest of two waves is called the wavelength which is typically measured in nanometers (nm). (1 nm=10^{-9} meter)
- Radiation can be differentiated according to its **wavelength**, **frequency** and / or **energy**, as the wavelength of the radiation **decreases** the energy emit by the radiation **increases**.
- Radiation has various effects on cells, depending on its wavelength, intensity, and duration.

Types of Radiation

1. **Ionizing radiation**

- Ionizing radiation has a wavelength that is typically **less than** (1) nm and energy that is **greater than** 100 electron Volts (eV).
- Ionizing radiation, such as x-rays or gamma radiation carries enough energy to remove electrons from molecules in a cell. When electrons are removed, ions called free radicals are formed. Free radicals can damage most other molecules in a cell, such as DNA or RNA by oxidizing them.
- Used to sterilize plastic petri dishes, surgical gloves, heat sensitive items and disposable medical supplies such as syringes.
- Used to sterilize meats, spices, and fresh fruits and vegetables.
- Disadvantages:
    - Continuous exposure to ionizing radiation does achieve sterility
    - Penetrates human tissues, may cause genetic mutations.

2. **Non-ionizing radiation**

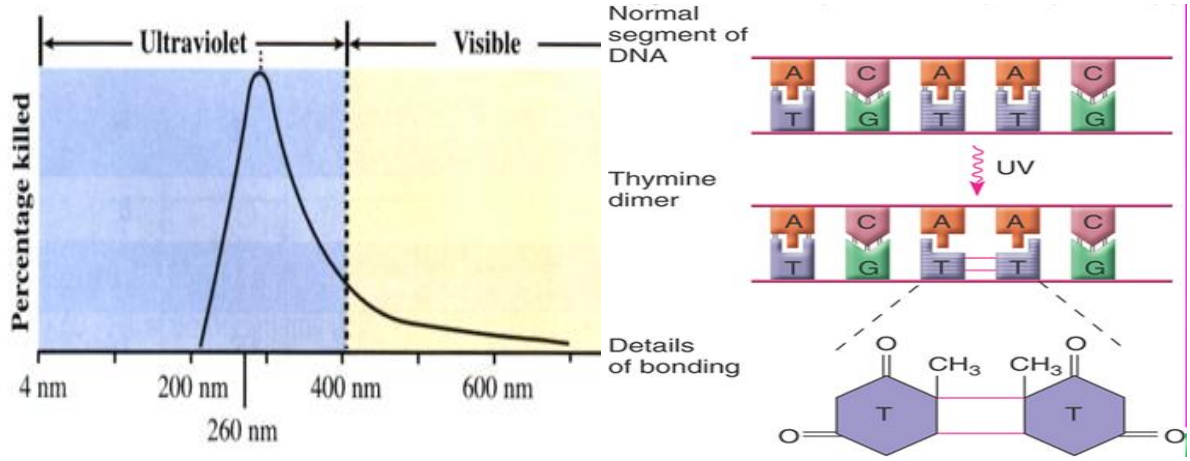
- Non-ionizing radiation has a wavelength that is **greater than 1** nm and energy that is **only a few to** 100 eV.
- Non-ionizing radiation such as Ultraviolet (UV) light excites electrons and causes them to make new covalent bonds but does not ionize the molecules.
- Used to reduce microbial populations on surfaces, heat sensitive items, hospital rooms, operating rooms.
- Disadvantages:
    - Damages skin and eyes.
    - Does not penetrate plastic or glass matters due to very low penetrating power.

Ultraviolet (UV) light

- Ultraviolet light is a kind of non-ionizing radiation with a wavelength from **100 nm to 400 nm**.
- Ultraviolet light is divided into three groups by wavelength, that have different degrees of harmful effects:
  1. **UV-A**: wavelengths ranging from 315 to 400 nm.
  2. **UV-B**: wavelengths between 280 and 315 nm.
  3. **UV-C**: wavelengths ranging from 100 to 280 nm.

Lethal effects of UV light

- UV of wavelength **260** nm is strongly absorbed by the bases in DNA which may lead to mutation or cell death by induce the formation of pyrimidine (particularly **thymine**) dimer which arises by the formation of covalent bonds between the 5,6-positions of two adjacent pyrimidine residues.
- Dimers block DNA replication and transcription.
- Non-spore formers = UV sensitive
- Endospore formers = UV resistant
  - *Bacillus subtilis*



Applications of UV light

Ultraviolet light is commonly used to disinfect the:

- Laboratory work surfaces.
- Eggs surfaces.
- Air and surfaces in hospital rooms.
- Vaccines and other medical products.

Aim

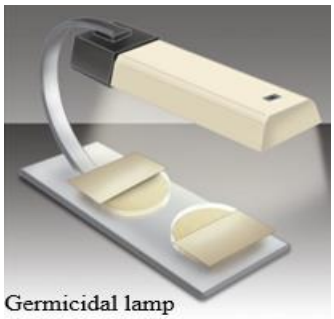
The purpose of this experiment is to determine the effects of short terms ultraviolet light exposure on growth of certain bacterial species.

Materials

- Nutrient agar plates
- Bacterial cultures
- Ultraviolet lamp
- Index cards

Procedure

1. Label 5 plates with bacteria name, exposure time.
2. Label ½ plate "Exposed" & other ½ plate "Unexposed".
3. Swab entire surface of plate agar with:
   - *E. coli*
   - *Staph. aureus*
   - *Bacillus subtilis*
4. Remove lid & cover unexposed ½ of plate with index card
   An open media plate culture is exposed to UV light. An index card is covering ½ of the plate.
5. Place plates under UV light
   - Group 1 = (1) minute
   - Group 2 = (3) minutes
   - Group 3 = (5) minutes
   - Group 4 = (7) minutes
   - Group 5 = (9) minutes


Germicidal lamp

6. Replace lid & incubate plates upside down at 37º C for 24 hours.

WARNING:

- Do not look directly to the UV light as it can cause damage to your eyes.
- Do not expose your skin directly to the UV, wear gloves.

Results

Observations and Interpretations

- Count the colonies on the side of the plates that were exposed to UV light and record the colonies number in the following data table.

Microorganism	**UV Exposure time**					
	Unexposed	**1 min**	**3 min**	**5 min**	**7 min**	**9 min**
Escherichia coli						
Staph. aureus						
Bacillus subtilis						