Chapter 6: Radiobiology

NPRE441: Principles of Radiation Protection
Spring 2023, MW 12-1.50 pm
2018 Campus Instructional Facility

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Objective:
To familiarize the students with the basic principles of radiobiology.

Slides retrieved and adapted from:
• Slide deck NPRE441 Spring 2021 by Prof. L.J. Meng (UIUC, USA)
• slide deck prepared in 2006 by Dr. E.B. Podgorsak (McGill University, Montreal)
• slide deck prepared in 2015 by Dr. M. Cremonesi (IEO European Institute of Oncology, Milano, Italy)
• slide deck prepared by Dr. E. Okuno (Institute of Physics of S. Paulo University, S. Paulo, Brazil)
CHAPTER 6.  

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PART 4

- 5 R’s of Radiobiology
  - Repopulation
  - Reoxygenation
  - Redistribution
  - Intrinsic Radiosensitivity
  - Repair
Electromagnetic radiation is considered ionizing if it has a photon energy greater than

1. 1.24 eV  
   [13.04%]
2. 12.4 eV  
   [39.13%]
3. 124 eV  
   [21.74%]  
   Correct Answer
4. 1.24 keV  
   [26.09%]
UV 3.10-12.4 eV
Near IR 0.12-1.77 eV
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Radiobiology (or radiation biology) is a branch of science which combines the basic principles of physics and biology and is concerned with the interaction of ionizing radiation on living matter.

(potential) biological effects produced by energy absorption in small volumes corresponding to single cells or parts of cells.
6.1. INTRODUCTION

2. International Organisations on Radiation effects

**BEIR**
(National Academy of Sciences, Biological Effects of Ionizing Radiation)

**UNSCEAR** (United Nations Scientific Committee on the Effects of Atomic Radiation)

**ICRP**
(International Commission on Radiological Protection)

- Collect and analyze data from the recent literature regarding biological effects of ionizing radiation

- Report periodically on risk estimates for radiation induced cancer and hereditary effects

- is involved in recommendation and development of guidelines in the field of radiation protection
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

- All living entities are made up of **protoplasm**, which consists of inorganic and organic compounds dissolved or suspended in **water**.

- The smallest unit of protoplasm capable of independent existence is the **cell**, the **basic microscopic unit of all living organisms**.
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

- Group of cells that together perform one or more functions is referred to as tissue.

- An organ is a collection of tissues that structurally form a functional unit specialized to perform a particular function.

- Group of organs that perform one or more functions is an organ system or an organism.
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

- Cells are separated from the surrounding environment by a cell membrane and contain:
  - Inorganic compounds (water and minerals)
  - Organic compounds (proteins, carbohydrates, nucleic acids, lipids)

- The two main constituents of a cell are:
  - Cytoplasm supports all metabolic functions within a cell.
  - Nucleus contains the genetic information (DNA).

- Cell components inside the cell membrane are called organelles: subcellular structures that have one or more specific functions in the cell.
  - nucleus stores the genetic information
  - mitochondria produce chemical energy
  - ribosomes assemble proteins, etc...

https://www.genome.gov/genetics-glossary
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

3D rendering (NOT direct image) of a eukaryotic cell that was modeled using X-ray, nuclear magnetic resonance (NMR), and cryo-electron microscopy datasets. From “Cellular landscape cross-section through a eukaryotic cell.” by Evan Ingersoll and Gael McGill (Nov 2020).

https://www.digizyme.com/cst_landscapes.html
A cell is the basic building block of living things. All cells can be sorted into one of two groups: eukaryotes and prokaryotes.
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

three-domain classification
### 6.1 INTRODUCTION

#### 3. Basic Concepts of Cell Biology

![Venn Diagram](https://vimeo.com/channel/s/1659505/152405826)

**PROKARYOTES**
- oldest cell type
- small and simple
- lack nucleus
- lack organelles
- single-celled
- single circular chromosome

**EUKARYOTES**
- evolved from prokaryotes
- larger and more complex
- contain nucleus
- contain organelles
- single-celled or multicellular
- multiple linear chromosomes

**BOTH**
- have DNA
- have ribosomes
- have cytoplasm
- have plasma membrane
6.1 INTRODUCTION
3. Basic Concepts of Cell Biology

Prokaryotic vs Eukaryotic

- Prokaryotic cells are referred to as simple and unicellular (single-celled) organisms
- They lack a nucleus, and do not have any membrane bound organelles
- The nucleoid of prokaryotic cells contains DNA that is small and circular
- Prokaryotic cells have less cell parts compared to Eukaryotic Cells, they are DNA, ribosomes, cell Membrane, cell Wall, there are other cell parts that are not present in all Prokaryotic Cells
- Some examples of Prokaryotic cells include Bacteria and Archaea

- Eukaryotic cells referred to as complex and multicellular organisms
- They have a nucleus as well as membrane-bound organelles within the cell.
- The DNA or genetic material of a Eukaryotic cell is found within the cell's nucleus.
- A plasma membrane, cytoplasm, nucleus, ribosomes, mitochondria, peroxisomes, and, in some cases, vacuoles make up a Eukaryotic cell. However, there is a distinction between animal and plants
- Examples of Eukaryotic cells include animals and plants
3. Basic Concepts of Cell Biology
Human cells are either somatic cells or germ cells. Germ cells are either a sperm or an egg, all other human cells are called somatic cells.
6.1 INTRODUCTION

3. Basic Concepts of Cell Biology

Mitosis occurs in somatic cells, cells that make up the body’s organs or tissues. Mitosis allows the body tissues to be repaired or to grow, and allows single-celled organisms to reproduce.

- The cell that divides is called the parent cell, while the resulting in two new identical daughter cells that have the same number of chromosomes as the parent cell.

Meiosis occurs in gametes or cells that are involved in sexual reproduction. Meiosis allows gametes (egg and sperm cells) to be created and allows multicellular organisms to reproduce.

- The cell divides into two cells, and then after the first division it divides again resulting in four cells with half the number of chromosomes as the parent cell.
Somatic cells are classified as:

- **Stem cells** (SCs), which exist to self-perpetuate and produce cells for a differentiated cell population.
- **Transit amplifying cells** (TACs) are an undifferentiated population in transition between SCs and differentiated cells.
- **Mature cells**, which are **fully differentiated** (have developed a specific morphology and function) and do not exhibit mitotic activity (static).
6.1 INTRODUCTION

4. Cell Cycle

- Time between successive divisions (mitoses) is called cell cycle time.

- **Cell cycle time for stem cells** in certain tissues is up to 10 days.

- Cell cycle time for mammalian cells is of the order of **10 – 20 hours** and is divided in **4 phases**:
  - **S phase** is usually in the range of 6 – 8 hours (50%).
  - **M phase** is less than 1 hour (5%).
  - **G₂** is in the range of 2 – 4 hours (15%).
  - **G₁** is in the range of 1 – 8 hours (30%).
  - **G₀** describes a cellular state outside of the replicative cell cycle.
6.1 INTRODUCTION

4. Cell Cycle

The Cell Cycle

- **G2 (2nd gap)**
  - Final preparation for mitosis phase. Cell provides energy (ATP) and synthesizes essential protein.

- **M (Mitosis)**
  - Cell nucleus divides. Pairs of chromatid separate from each other.

- **Cytokinesis**
  - Cytoplasm divides to form two new cells.

- **S (Synthesis of DNA)**
  - Cell starts replicating chromosomal DNA. Results in pairs of chromatid at centrometric region.

- **G1 (1st gap)**
  - Cell grows and accumulates essential protein as DNA building block.
Cell proliferation cycle is defined by two time periods:

- **Mitosis M**, where division takes place.
- The period of DNA synthesis **S**.

S and M portions of the cell cycle are separated by two periods (gaps) **G₁** and **G₂** when, respectively

- DNA has not yet been synthesized.
- Has been synthesized but other metabolic processes are taking place.
Chromosome is a microscopic, threadlike part of a cell that carries hereditary information in the form of genes.

Every species has a characteristic number of chromosomes; humans have 23 pairs (22 pairs are non-sex chromosomes and 1 pair is sex chromosome).

Gene is a unit of heredity that occupies a fixed position on a chromosome.
Deoxyribonucleic acid (DNA) contains the genetic information of the cell

- DNA is a large molecule and has a characteristic double-helix structure (twisted ladder) consisting of two strands, each made up of a sequence of nucleotides.
- The backbone of the DNA strand is made of alternating sugar and phosphate groups.
- A nucleotide is a subunit of DNA, and is composed of a base linked to a sugar (deoxyribose) and a phosphate group.
- Each cell contains about 2m of DNA.

J. D. WATSON and F. H. C. CRICK
The **four bases of DNA** can be classified in two groups:

- **Purines**
  - adenine (A)
  - guanine (G)

- **Pyrimidines**
  - cytosine (C)
  - thymidine (T)

- The unique pairing of the nucleotide bases provides DNA with its identity which is used in replication.
- One of the pair must be a purine and the other a pyrimidine for bonding to occur.
- The cell’s genetic information is carried in a linear sequence of nucleotides that make up the organism’s set of genes.
6.1 INTRODUCTION

5. Structure of DNA

DNA molecules are made up of two strands that wrap around each other, forming something that looks like a ladder! This shape is called a double helix.

The backbone of the DNA molecule is made up of the sugar and phosphate groups of the nucleotides, which are joined together by phosphodiester bonds.

The inside portion of the DNA molecule, or the rungs of the ladder, each consist of 2 nitrogenous bases from either strand, which are hydrogen bonded to each other.

Each DNA strand is made up of many nucleotides linked together in a chain. Every nucleotide consists of a sugar (deoxyribose), a phosphate group, and a nitrogenous base.

DNA nucleotides can contain one of 4 different nitrogenous bases: adenine (A), thymine (T), guanine (G), or cytosine (C).

There are specific rules for the pairing of nitrogenous bases! Adenine only pairs with thymine (A-T), and cytosine only pairs with guanine (C-G).
The main constituents of a cell are:

1. cytoplasm 82.14%
2. organelles such as mitochondria, ribosomes, etc... 92.86%
3. protoplasm 14.29%
4. nucleus 96.43%
5. meiosis 0%

(% = Percentage of Voters)
About Somatic cells:

Vote for up to 2 choices

1. Their cellular division process is called meiosis and results in two genetically identical daughter cells
   - 7.14%

2. can only be prokaryote cells
   - 67.86%

✓ 3. can be stem cells, transit amplifying cells, mature cells
   - 64.29%

✓ 4. Are complex cells characterized by cytoplasm, nucleus and several organelles
   - 57.14%

(% = Percentage of Voters)
The meiosis:

1. Is the division of germ cells that involves two fissions of the nucleus giving rise to four sex cells, each possessing half the number of chromosomes of the original germ cell
   - 96.43%
2. Is typical of prokaryote cells
   - 10.71%
3. Is one of the phases in a cell cycle (M)
   - 50%
4. Is cellular state outside of the replicative cell cycle
   - 28.57%

(% = Percentage of Voters)
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6.2 IRRADIATION OF CELLS

1. Time Frame of Radiation Effect

When cells are exposed to ionizing radiation:

- First, the standard physical effects between radiation and the atoms or molecules of the cells occur.
- Possible biological damage to cell functions follows.
6.2 IRRADIATION OF CELLS

1. Time Frame of Radiation Effect: PHYSICAL STAGE

- Radiation interacts with molecules and atoms: mainly water, since about 80% of a cell is composed of water to produce free radicals.

- A free radical is a molecule or atom, which is not combined to anything (i.e., free) and carries an unpaired electron in its outer shell, i.e. it’s looking for something to interact with, or in purely scientific terms, it is in a state associated with a high degree of chemical reactivity.

- Basic radiochemical reactions in the physical stage are:

  1. \( H_2O \xrightarrow{h\nu} H_2O^+ + e^- \)
  2. \( H_2O \xrightarrow{h\nu} H_2O^* \)

The initial changes produced by radiation in water are the creation of (1) ionized and (2) excited molecules. They are produced in \(<10^{-15}s\) in local regions of the track.
Prechemical Stage

- The time between $10^{-15}$s to $10^{-12}$s are called the **pre-chemical stage**.
- The species produced by the radiation ($\text{H}_2\text{O}^+$, $\text{H}_2\text{O}^*$ and free, sub-excitation electrons) induce chemical reactions as follows:

**Process #1:**
An ionized water molecule reacts with a neighboring molecule, forming a **hydronium ion** and a **hydroxyl radical**:

$$\text{H}_2\text{O}^+ + \text{H}_2\text{O} \rightarrow \text{H}_3\text{O}^+ + \text{OH}.$$  

**Process #2**
An excited water molecule gets rid of its energy either by losing an electron, thus becoming an ion radical and proceeding according to the reaction or by **molecular dissociation**:

$$\text{H}_2\text{O}^* \rightarrow \begin{cases} \text{H}_2\text{O}^+ + \text{e}^- \\ \text{H} + \text{OH} \end{cases}$$
Prechemical Changes in Irradiated Water

Process #3:

- The sub-excitation free electrons migrate, losing energy by vibrational and rotational excitation of water molecules, and become thermalized by times $\sim 10^{-12}$ s.

- The thermalized electrons orient the permanent moments of neighboring water molecules, forming a cluster, called a hydrated electron:

$$e^- \rightarrow e_{aq}^-$$

Active radiolytic species

- end products of the pre-chemical stage

\[ H_3O^+ , \quad OH , \quad e_{aq}^- , \quad \text{and} \quad H \]

- Hydronium ion
- Hydroxyl
- Hydrated electron

Radiolysis is the dissociation of molecules by ionizing radiation.
Chemical Stage

- The time between $10^{-12}$s to $10^{-6}$s are called the chemical stage.
- The chemical stage is characterized by diffusion-controlled reactions.
- The average distance a particle traveled $\lambda$ is related to time $\tau$ and the diffusion constant $D$ by

$$\frac{\lambda^2}{6\tau} = D$$

- Two species that are closer than the sum of their reaction radii will have a chance to interact.
FIGURE 13.1. Chemical development of a 4-keV electron track in liquid water, calculated by Monte Carlo simulation. Each dot in these stereo views gives the location of one of the active radiolytic species, OH, H$_3$O$^+$, e$_{aq}^-$, or H, at the times shown. Note structure of track with spurs, or clusters of species, at early times. After $10^{-7}$ s, remaining species continue to diffuse further apart, with relatively few additional chemical reactions. (Courtesy Oak Ridge National Laboratory, operated by Martin Marietta Energy Systems, Inc., for the Department of Energy.)
**Example**

Estimate how far a hydroxyl radical will diffuse in $10^{-12}$ s.

**Solution**

From Eq. (13.11) with $\tau = 10^{-12}$ s and from Table 13.2, we find

$$\lambda = (6\tau D)^{1/2} = (6 \times 10^{-12} \text{ s} \times 2 \times 10^{-5} \text{ cm}^2 \text{ s}^{-1})^{1/2}$$

$$= 1.10 \times 10^{-8} \text{ cm} = 1.10 \text{ Å}. \quad (13.12)$$

For comparison, the diameter of the water molecule is 2.9 Å. The answer (13.12) is compatible with our taking the time $\sim 10^{-12}$ s as marking the beginning of the chemical stage of charged-particle track development.

**Table 13.2** Diffusion Constants $D$ and Reaction Radii $R$ for Reactive Species

<table>
<thead>
<tr>
<th>Species</th>
<th>$D \ (10^{-5} \text{ cm}^2 \text{ s}^{-1})$</th>
<th>$R \ (\text{Å})$</th>
</tr>
</thead>
<tbody>
<tr>
<td>OH</td>
<td>2</td>
<td>2.4</td>
</tr>
<tr>
<td>$e^{-}_{aq}$</td>
<td>5</td>
<td>2.1</td>
</tr>
<tr>
<td>$H_3O^+$</td>
<td>8</td>
<td>0.30</td>
</tr>
<tr>
<td>H</td>
<td>8</td>
<td>0.42</td>
</tr>
</tbody>
</table>

Turner, pp. 402.
Chemical Stage

After $\sim 10^{-12}$ s, the four chemically active species $\text{H}_2\text{O}^+$, $\text{OH}$, $e^-_{\text{aq}}$, and $\text{H}$ are located near the positions of the original $\text{H}_2\text{O}^+$, $\text{H}_2\text{O}^*$, and $e^-$ that triggered their formation.

Three of the new reactants, $\text{OH}$, $e^-_{\text{aq}}$, and $\text{H}$ are free radicals. They begin to migrate in thermal motion. Individual pairs of these reactants may get sufficiently close to induce chemical reactions, for example

\[
\begin{align*}
\text{OH} + \text{OH} &\rightarrow \text{H}_2\text{O}_2, \\
\text{OH} + e^-_{\text{aq}} &\rightarrow \text{OH}^-, \\
\text{OH} + \text{H} &\rightarrow \text{H}_2\text{O}, \\
\text{H}_3\text{O}^+ + e^-_{\text{aq}} &\rightarrow \text{H} + \text{H}_2\text{O}, \\
e^-_{\text{aq}} + e^-_{\text{aq}} + 2\text{H}_2\text{O} &\rightarrow \text{H}_2 + 2\text{OH}^- , \\
e^-_{\text{aq}} + \text{H} + \text{H}_2\text{O} &\rightarrow \text{H}_2 + \text{OH}^- , \\
\text{H} + \text{H} &\rightarrow \text{H}_2.
\end{align*}
\]
Chemical Stage

- As time passes, the reactions proceed until the remaining reactants diffused so far away from one another that the **probability of additional chemical reaction becomes very small**.

- These happen typically within $10^{-6}$ s, by which the chemical development of the track in pure water is essentially over.
Select the chemical reactions happening during the physical stage:

Vote for up to 2 choices

1. a. $e^- \rightarrow e^-_{aq}$
   - 17.86%

2. b. $H_2O \xrightarrow{hv} H_2O^+ + e^-$
   - 75%

3. c. $H_2O^+ + H_2O \rightarrow H_3O^+ + OH.$
   - 28.57%

4. d. $H_2O \xrightarrow{hv} H_2O^*$
   - 67.86%

(\% = Percentage of Voters)
Which of the following statements concerning free radicals is TRUE?

1. Free radicals can be generated only by gamma radiation but not X-rays  
   0%
2. Free radicals have half-lives on the order of seconds  
   18.52%
3. Free radicals carry a net electrical charge, and therefore a low degree of chemical reactivity  
   3.7%
4. Free radicals have an unpaired electron in the outer shell  
   77.78%
The lifetime of radicals in target molecules is about:

1. $10^{-3}$ sec  18.52%
2. $10^{-6}$ sec  25.93%
3. $10^{-9}$ sec  22.22%
4. $10^{-12}$ sec  33.33%
Lapse of Time after Exposure and Effects

- Physio-chemical processes
- Biochemical process
- Biological process
- Clinical process

Physiological processes:
- Physical
- Prechemical
- Chemical

Stages:
- One-thousandth of a second after irradiation
- Minutes-days
- Weeks-Years

Effects:
- Stochastic effects: Cancer, Hereditary effects
- Deterministic effects: Acute radiation syndromes, Fetal effects, Cataract
Interaction, Damage & Repair of DNA

(1) DNA Interaction

(2) Damage

(3) Repair

Complete repair

Partial or incomplete repair

Ministry of the Environment
Government of Japan
6.2 IRRADIATION OF CELLS

2. Interaction with DNA

- **Sensitive component** for radiation-induced cell killing rests in the cell nucleus (DNA) and **NOT** in the cytoplasm.

- When directly ionizing radiation is absorbed in biological material, the damage to the cell may occur in one of two mechanisms:
  - Direct
  - Indirect

**Diagram:** Difference between direct and indirect damage to cellular DNA
6.2 IRRADIATION OF CELLS

2.1 Direct action

- In direct action the radiation interacts directly with the critical target in the cell.
- Accounts for $\sim1/3$ of the damage.
- Direct action is the dominant process in the interaction of high LET particles such as neutrons or alpha particles with biological material.
- In direct action caused by x-ray or gamma ray photons, the photon interaction with an atom in the cell produces a charged particle (electron or positron) which subsequently interacts with the DNA directly.

1 Gy $\sim 3 - 4$ tracks per cell

1 Gy $\sim 1000$ tracks per cell
In indirect action the radiation interacts with other molecules and atoms (mainly water, since about 80% of a cell is composed of water) within the cell to produce free radicals, which can, through diffusion in the cell, interact and damage the critical target within the cell.

accounts for ~2/3 of the damage.

Basic radiochemical reactions that may occur in water molecules disrupted by passage of an ionizing particle are as follows:

1. \( \text{H}_2\text{O} \rightarrow \text{H}_2\text{O}^+ + e^- \rightarrow \text{H}_2\text{O}^+ + e_{aq}^- \)

2. \( \text{H}_2\text{O}^+ \rightarrow \text{OH}^+ + \text{H}^+ \)  
   75% indirect damage

3. \( \text{H}_2\text{O} \rightarrow \text{H}_2\text{O}^* \rightarrow \text{H}^* + \text{OH}^* \)  
   Reactive Oxygen Species (ROS) (due to an unpaired valence electron) produced in water. Because short life of simple free radicals, only those formed in water column of 2-3 nm around DNA are able to participate in indirect effect.
Radiation induced damage to biological targets may result from:

**Direct action**
- Predominant with high LET radiation, e.g. $\alpha$, neutrons
- Ionization or excitation (via Coulomb interactions) of the atoms in the **biological target** (for example direct break-up of DNA molecules)
- In normally oxygenated mammalian cells, direct effects account for $\sim 1/3$ of the damage for low LET radiations such as electrons and photons.

**Indirect action**
- Predominant with low LET radiation, e.g. $X$, $\gamma$ rays
- Radiation effects on atoms or molecules which are NOT parts of the biological target.
- Cells exist in a rich aqueous environment → the majority of indirect actions involve the ionization or excitation of water molecules. The free radicals created may then migrate and damage the adjacent biological targets.
- Indirect action is the main cause of radiation damage and, in normally normoxic cells, accounts for $\sim 2/3$ of the damage.
2.2 Indirect action in cell damage by radiation

Steps involved in producing biological damage by the indirect action of x rays are as follows:

- **Primary photon interaction** (photoelectric effect, Compton effect, pair production) produces a high energy electron, ion radicals or excited molecules.

- **High energy charged particle** in moving through tissue produces free radicals in water.

- **Free radicals** may produce chemical changes in DNA from the breakage of chemical bonds.

- **Changes in chemical bonds** result in biological effects.
Through a Compton scattering event with water, a photon will produce: 1) a free electron with high energy, 2) an ionized water molecule, and 3) the original photon with reduced energy $<124$ eV. Which of the following subsequent reactions is considered an INDIRECT effect of the original photon?

1. The high-energy free electron further interacts with a different water molecule to cause another ionization event
   - 7.14%

2. The ionized water molecule encounters another water molecule, and produces a hydroxyl free radical
   - 35.71%

3. The original photon continues to ionize other water molecules
   - 7.14%

4. All of the above
   - 50%
Explanation:

The interaction of ionized water molecule with another water molecule produces new species of free radicals and it is considered the indirect effect. The newly produced electron slows down to form an aqueous electron (surrounded by a cluster of polarized water molecules), it can interact with other water molecules to trigger indirect effects. The remaining photon has an energy lower than the minimum energy required for ionization, therefore it does not contribute anymore to indirect effects.
Which of the following is NOT a characteristic of the INDIRECT action of ionizing radiation

1. Production of diffusible free radicals
   - 7.69%

2. Production of reactive oxygen species
   - 3.85%

3. Accounts of 1/3 of the radiation damage
   - 61.54%

4. Dominant process in the interaction of low LET radiation
   - 26.92%
### 6.2 IRRADIATION OF CELLS

#### 3. DNA damage

- DNA damage is the primary cause of cell death induced by radiation.
- Radiation exposure produces a wide range of lesions in the DNA:

1. **Base damage**
   - 2500 locations
   - Repaired in 5-10 min
2. **Single-strand break (SSB)**
   - 1000 locations
   - Repaired in 10-20 min
3. **Double-strand break (DSB)**
   - 30-40 locations
   - Repaired in >50 min

---

Source: Morgan, Annual Meeting of the National Committee on Radiation Protection and Measurements (NCRP) (44th, 2008)
At lower doses and dose rates (multiple exposures), cellular recovery may play an important role in the fixation of the radiation damage.

There are three broad types of cellular radiation damage:

- **Lethal damage**: in which the cellular DNA is irreversibly damaged to such an extent that the cell dies or loses its proliferative capacity.

- **Sublethal damage**: in which partially damaged DNA is left with sufficient capacity to restore itself over a period of a few hours, provided there is no further damage during the repair period.

- **Potentially lethal damage**: in which repair of what would normally be a lethal event is made possible by manipulation of the post-irradiation cellular environment (cells are allowed to remain in the non-dividing state $G_0$).
Sub-lethal (or accumulated) damage results from accumulation of events that individually are incapable of killing a cell but that together can be lethal.
4. DNA repair

The number of DNA lesions generated by irradiation is large, but there are a number of mechanisms for DNA repair → the **percentage of lesions causing cell death is very small**

DNA repair mechanisms

Important for the recovery of cells from radiation and other damaging agents.

There are **multiple enzymatic mechanisms** for detecting and repairing radiation induced DNA damage.

DNA repair mechanisms:
- base excision repair
- mismatch repair
- nucleotide excision repair
- Non-homologous end joining (NHEJ)
- Homologous recombination
- Double-strand repair
6.2 IRRADIATION OF CELLS

4. DNA repair

Error-free repair

Faulty repair

No or failed repair

The damage is totally removed

The damage causes mutations not lethal or lethal but in the long-term

The damage is lethal for the cell

Cell survival

Neoplasia

Cell death
6.2 IRRADIATION OF CELLS

Radiosensitivity differs throughout the cell cycle with, in general:

- late S phase being the most radioresistant
- G₂/M being the most radiosensitive (Cells going through the division phase)
- G₁ phase taking an intermediate position

- The greater proportion of DNA enzymatic repair during late S phase may explain the resistance of late S phase cells
- Poor repair competence (reduced enzyme access due to chromatin compaction) explains the high radiosensitivity in G₂/M phase
- Resting cells in G₀, not involved in the cell cycle, are more resistant to radiation when compared to late S-phase cells
Differentiating Cells: these cells are sensitive to radiation; they are relatively short-lived and include the first generation produced by division of the vegetative mitotic cells.

Totally Differentiated Cells: these cells are relatively radioresistant; they normally have relatively long lifespans and do not undergo regular or periodic division in the adult stage, except under abnormal conditions such as following damage to or destruction of a large number of their own kind.

Fixed Nonreplicating Cells: these cells are most radioresistant; they are highly differentiated morphologically and highly specialized in function.
Possible outcomes of cell irradiation:

- **No effect.**
- **Division delay:** The cell is delayed in going through division.
- **Apoptosis:** The cell dies before it can divide.
- **Reproductive failure:** The cell dies when attempting the mitosis.
- **Mutation:** The cell survives but contains a mutation (transformed genotype).
- **Transformation:** The mutation leads to a transformed phenotype and possibly carcinogenesis.
- **Bystander effects:** An irradiated cell may send signals to neighboring unirradiated cells and induce genetic damage in them.
- **Adaptive responses:** The irradiated cell becomes more radio-resistant.
Radiation doses of the order of several Gy may lead to cell loss. Cells are regarded as having been ‘killed’ by radiation if they have lost reproductive integrity, according to the following mechanisms:

- **Apoptosis** or **programmed cell death** can occur naturally or result from insult to the cell environment. It occurs in particular cell types after low doses of irradiation.

- **Necrosis** is a form of cell death associated with loss of cellular membrane activity. Cellular necrosis generally occurs after high radiation doses.

- **Mitotic catastrophe** involves cells attempting to divide without proper repair of DNA damage leading to a reproductive cell death which can occur in the first few cell divisions after irradiation, and with increasing frequency after increasing doses.

- **Senescence** Senescent cells are metabolically active but have lost the ability to divide.
Surviving cell that maintains its reproductive integrity and proliferates almost indefinitely into a large number of progeny is said to be clonogenic.

Capability of a single cell to grow into a large colony shows that the cell has retained its reproductive integrity.

In general, to destroy cell function in non-proliferating mature (static) cells a typical dose of 100 Gy is required, while to destroy proliferative cells (stem cells) requires typically only 2 Gy.

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**METABOLIC DEATH**

**REPRODUCTIVE DEATH**
Approximately how many DNA double strand breaks (DSB) are caused per cell per Gray?

1. 1-10
   - 8.7%
2. 30-40
   - 73.91%
3. 1000
   - 8.7%
4. 2500
   - 8.7%

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The lethal lesion caused in DNA by low LET ionizing radiation

1. Can be restored over a period of several hours through different enzymatic mechanisms  
   - 20%

2. Is caused by alpha-type events  
   - 0%

3. Does not correlate with the cell proliferative capacity  
   - 0%

4. Is due to the accumulation of multiple DNA double-strand breaks  
   - 80%
Which of the following is correct about alpha-type cell killing following radiation exposure

1. It represents single lethal hits
   - 30.43%
2. can be avoided only if the cell to go in the "hibernation" state G0
   - 17.39%
3. It accounts for >60% of the damage
   - 39.13%
4. It is oxygen dependent
   - 13.04%
Radiation Effect on Biological Systems

(1) CELL IRRADIATION

DIRECT ACTION: IONIZATION OF DNA

INDIRECT ACTION: IONIZATION OF OTHER MOLECULES, e.g., H₂O

\[ \text{radiation} + \text{H}_2\text{O} \rightarrow \text{H}_2\text{O}^+ + e^- \]

\[ H_2O^+ \rightarrow H^+ + O\text{H}^0 \]

\[ e^- + H_2O \rightarrow H^0 + O\text{H}^- \]

OXIDATION OF DNA BY OH RADICALS

(2) DNA DAMAGE:

- Lethal
- Sublethal
- Potentially lethal

(3) ENZYMATIC REPAIR

DNA RESTORED

NO EFFECT

CELL DEATH:

1) Metabolic for mature cells
2) Reproductive for stem cells

PERMANENT DAMAGE IN DNA

BIOLOGICAL EFFECTS

1. GENETIC EFFECTS vs SOMATIC EFFECTS
2. STOCHASTIC vs DETERMINISTIC EFFECTS
3. ACUTE vs LATE EFFECTS
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