Presented at the
Armed Forces Radiobiology Research Institute
Scientific Medical Effects of Ionizing Radiation Course
July 28 through August 1, 2008
Bethesda, Maryland

Distributed via the AFRRI Web site
http://www.afrri.usuhs.mil

The Scientific Medical Effects of Ionizing Radiation Course, conducted once a year, focuses on the latest research about the medical effects of ionizing radiation to help clinicians, health physicists, and medical planners preserve troop health in the face of radiological/nuclear terrorism or warfare.

For additional information about AFRRI training opportunities, contact AFRRI Military Medical Operations at 301-295-9150 or press the "Request info about: MEIR courses" button on this web page. To view more AFRRI information products, go to this web page.

For questions or more information about the content of this presentation, contact the presentation author.
**Fundamentals of Radiation Biology**

**Report Documentation Page**

<table>
<thead>
<tr>
<th>1. REPORT DATE</th>
<th>JUL 2008</th>
</tr>
</thead>
<tbody>
<tr>
<td>2. REPORT TYPE</td>
<td></td>
</tr>
<tr>
<td>3. DATES COVERED</td>
<td>00-00-2008 to 00-00-2008</td>
</tr>
<tr>
<td>4. TITLE AND SUBTITLE</td>
<td>Fundamentals of Radiation Biology</td>
</tr>
<tr>
<td>5a. CONTRACT NUMBER</td>
<td></td>
</tr>
<tr>
<td>5b. GRANT NUMBER</td>
<td></td>
</tr>
<tr>
<td>5c. PROGRAM ELEMENT NUMBER</td>
<td></td>
</tr>
<tr>
<td>5d. PROJECT NUMBER</td>
<td></td>
</tr>
<tr>
<td>5e. TASK NUMBER</td>
<td></td>
</tr>
<tr>
<td>5f. WORK UNIT NUMBER</td>
<td></td>
</tr>
<tr>
<td>7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)</td>
<td>Uniformed Services University of the Health Sciences, Armed Forces Radiobiology Research Institute (AFRRI), 8901 Wisconsin Avenue, BG 42, Bethesda, MD, 20889-5603</td>
</tr>
<tr>
<td>8. PERFORMING ORGANIZATION REPORT NUMBER</td>
<td></td>
</tr>
<tr>
<td>9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)</td>
<td></td>
</tr>
<tr>
<td>10. SPONSOR/MONITOR’S ACRONYM(S)</td>
<td></td>
</tr>
<tr>
<td>11. SPONSOR/MONITOR’S REPORT NUMBER(S)</td>
<td></td>
</tr>
<tr>
<td>12. DISTRIBUTION/AVAILABILITY STATEMENT</td>
<td>Approved for public release; distribution unlimited</td>
</tr>
<tr>
<td>13. SUPPLEMENTARY NOTES</td>
<td></td>
</tr>
<tr>
<td>14. ABSTRACT</td>
<td></td>
</tr>
<tr>
<td>15. SUBJECT TERMS</td>
<td></td>
</tr>
<tr>
<td>16. SECURITY CLASSIFICATION OF:</td>
<td></td>
</tr>
<tr>
<td>a. REPORT</td>
<td>unclassified</td>
</tr>
<tr>
<td>b. ABSTRACT</td>
<td>unclassified</td>
</tr>
<tr>
<td>c. THIS PAGE</td>
<td>unclassified</td>
</tr>
<tr>
<td>17. LIMITATION OF ABSTRACT</td>
<td>Same as Report (SAR)</td>
</tr>
<tr>
<td>18. NUMBER OF PAGES</td>
<td>27</td>
</tr>
<tr>
<td>19a. NAME OF RESPONSIBLE PERSON</td>
<td></td>
</tr>
</tbody>
</table>

*Standard Form 298 (Rev. 8-98)*

Prepared by ANSI Std Z39-18
Fundamentals of Radiation Biology

Scientific MEIR
AFRRI – July 2008
Col Mark S. Smyczynski
Objectives

- Describe chemistry of radiation absorption
- Describe cell survival curves and assay systems
- Describe interaction of ionizing radiation at cellular, tissue, and entire organism level
- Describe effect of dose rate
- Describe effect of time, dose, and fractionation
- Describe early and late reacting tissue response
- Describe acute effects of whole body radiation
- Describe oncogenic transformation 2° to radiation
Radiochemical reactions

incident photon
↓
fast electron $10^{-15}$ sec
↓
ion radical $10^{-10}$ sec
↓
free radical $10^{-5}$ sec
↓
breakage of chemical bonds $< 1$ sec
↓
biological effects hours to years
Direct & Indirect Action of Radiation

- Direct action:
  - Direct ionization of target
  - Secondary e\(^-\) directly ionizes target

- Indirect action:
  - Secondary e\(^-\) produces ion radicals that ionize target
  - Ion radicals produce free radicals that ionize target

- Indirect action predominates at ≈ 2:1
- Water commonly ionized as cell is 80% water
- Evidence supports DNA as the critical target
- More recent evidence demonstrates “bystander effect”
  - Likely related to release of cytotoxic agents, presence of gap-junctions, and membrane damage
Radiolysis of Water (Saline)

- \[ \text{H}_2\text{O} \rightarrow \text{H}_2\text{O}^+ + e^- \text{ (solvated electron)} \]
- \[ \text{H}_2\text{O}^+ + \text{H}_2\text{O} \rightarrow \text{H}_3\text{O}^+ + \text{OH}^- \text{ (hydroxyl radical)} \]
- \[ 2 \text{OH}^- \rightarrow \text{H}_2\text{O}_2 \text{ (hydrogen peroxide)} \]
- \[ e^- + \text{O}_2 \rightarrow \text{O}_2^- \text{ (dioxygen radical anion)} \]
- \[ \text{OH}^- + \text{alkyl (R)} \rightarrow \text{ROH}^- \text{ (alkyl free radical)} \]
- \[ \text{OH}^- + \text{Cl}^- \rightarrow \text{ClO}^- \text{ (hypochlorite anion)} \]
Cell Survival Curves

![Cell Survival Curves Diagram]

- **Survival**
- **αD**
- **βD'**
- **D1**
- **D0**
- **Dq**
- **Densely Ionizing (neutrons or α-rays)**
- **Sparsely Ionizing x-rays**
Linear Energy Transfer

Low LET
(photons)
Source: AFRRI

High LET
(alpha particles)
Source: AFRRI
Cell Survival Curves

- Refer to article (p 260-261) for more complete review
- Surviving fraction per linear-quadratic model

\[ \frac{S}{S_0} = e^{-\alpha D - \beta D^2} \]

\[ \frac{S}{S_0} = e^{-\alpha/\beta D - D^2} \]

\[ \frac{S}{S_0} = e^{-(\alpha/\beta D + D^2)} \]

- Significance of the $\alpha/\beta$ ratio covered subsequently
Radiobiology Assay Systems

- Cell survival curves represent *in vitro* conditions
  \[ S/S_0 = \frac{\text{colonies counted}}{(\text{cells seeded})(\text{PE/100})} \]
  where \( \text{PE} \) is defined as the plating efficiency
  \[ \text{PE} = \frac{\text{cells seeded}}{\text{cells that grow into colonies}} \]

- Clonogenic end point assays determined by observing a clone of regenerating cells *in situ*
  → murine skin colony assay
  → murine jujunal crypt cell assay
  → murine testes stem cell assay
  → murine kidney tubule assay
Radiobiology Assay Systems

- Clonogenic assays from donor animals
  eg: bone marrow stem cell assay
    (sometimes called spleen colony assay)
  step 1: lethally irradiate recipient mouse
  step 2: radiate donor mouse to test dose
  step 3: harvest bone marrow cells from donor mouse, form cell suspension, and inject into recipient mouse
  step 4: harvest spleen from recipient mouse 10 days later and count colonies
  \[
  S/S_0 = \text{colonies counted/cells inoculated} \times \text{PE}
  \]
Radiobiology Tumor System Assays

- Growth delay assay
  Radiate tumor and measure the time for regrowth to size at time of radiation or time to specified size

- TCD$_{50}$ assay (TDC = tumor control dose)
  Radiate tumors of uniform size at graded doses in series of animals, measure proportion controlled, and score dose achieving 50% local control

- Lung colony assay
  Radiate tumor to test dose, excise tumor, form cell suspension, inject into recipient mouse, harvest lungs 21 days later and count lung colonies
Radiosensitivity in the Mitotic Cycle

- Cell cycle: G1 → S → G2 → M → G1 etc.
  - Recall cells can enter into and out of G0 from G1
- Time for M almost universally at 1 hour
- Time for G2 quite consistent at 3 to 4 hours
- Time for S usually 6 to 8 hours and not > 15 hours
- Time for G1 highly variable from 1 to > 12 hours
- Mitotic harvest technique
- Synchronized cells obtained by block at end of G1
  - Cells accumulate at block using hydroxyurea then progress through cell cycle when drug removed
- Refer to article (p 261) regarding cell survival curves
Lethal damage
- Occurs subsequent to cytocidal radiation dose
- Damage irreversible and irreparable
- Most cells die in association with mitosis*
- Cell death usually occurs in subsequent mitosis
- Cells that die mitotic death may require up to 5 mitoses
- Some cells die from activated apoptotic pathways
- Many cell populations die both mitotic and apoptotic
- Radiosensitive cells tend to die from apoptosis
* Lymphocytes and oocytes die an interphase death
Classification of Radiation Damage

- Potentially lethal damage (PLD)
  Cytocidal under normal growth conditions
  Cell survival enhanced by modifying the post-irradiation cellular environment
  Suboptimal growth conditions inhibit cell cycle progression and complex process of mitosis
  Evidence indicates that PLD equates to DNA repair
Classification of Radiation Damage

- Sublethal damage (SLD)
  - Cell survival enhanced if total dose is divided in time
  - Two different patterns of repair demonstrated
  - Two fraction split dose experiments at 24°C & 37°C
  - One pattern of SLD repair demonstrated at 24°C when cells do not progress through the cell cycle
  - More complex pattern of SLD repair shown at 37°C
  - Prompt repair of SLD seen in first few hours
  - Surviving fraction decreases reaching low at 5 hours
  - Surviving fraction then increases again
Four R’s of Radiobiology

- Pattern of SLD repair based on mitotic cycle
- Three simultaneous processes account for pattern
  Prompt repair of SLD occurs initially
  In asynchronous population most sensitive cells die
  Surviving population of becomes partly synchronized
  Radioresistant S-phase cells progress through cycle
  Cell cycle progression often termed reassortment
  Cell division of surviving fraction causes repopulation
  First three “R’s” = repair - reassortment - repopulation
  Fourth “R” = reoxygenation represents separate topic
Dose Rate Effect

- Effect of dose rate extremely important
- Biologic effects strongly dependent on dose rate
- Dose rate effect essentially due to SLD repair
- Effect of dose rate separate from fractionation
- Refer to single page handout
Time, dose, & fractionation important in radiotherapy

- Time refers to the total time in days radiation delivered
- Dose refers to the total dose delivered
- Fractionation refers to the dose delivered per fraction

Conventional fractionation = 1.8 to 2.0 Gy/day

For a dose known to control a given burden of tumor at conventional fractionation, that dose must be increased when the standard treatment time exceeded

eg: 60 Gy over six weeks (thirty 2Gy/day fractions) does not have the same biological endpoint as 60 Gy over ten weeks while 80 Gy over ten weeks **might**
Fractionated Cell Survival Curves

![Graph showing fractionated cell survival curves for x-rays and neutrons with different RBE values. The graph plots surviving fraction against dose (cGy) on a logarithmic scale.]
RBE and OER

- Relative biological effectiveness = $D_{250kV_p}/D_{Test\ Radiation}$
  - required for equivalent biological effect
- $250\ kV_p$ x-rays “traditional” historic standard
- Numerical value of RBE dependent on isoeffect endpoint and can vary based on the TDF
- Oxygen enhancement ratio = $D_{Hypoxic}/D_{Aerated}$
  - required for equivalent biological effect
- Numerical value of OER dependent on isoeffect endpoint and can vary based on the TDF
- OER and reoxygenation only pertinent to radiotherapy
Early & Late Reacting Tissues

- At least two different tissue types recognized
- Early reacting tissues: actively mitotic
  e.g., skin & mucosa (buccal, intestinal, bladder)
- Late reacting tissues: post-mitotic
  e.g., connective tissue, bone, muscle, & nerve
- In linear-quadratic model, components of cell killing proportional to dose and \((dose)^2\) are equal when \(\alpha D = \beta D^2\) or \(D = \alpha/\beta\)
- The \(\alpha/\beta\) ratio defines the type of tissue response
- Early reacting: \(\alpha/\beta \approx 10\) Gy; late reacting: \(\alpha/\beta \approx 2\) Gy
- Shape of cell survival curve differ (refer to figure)
- Volume of tissue irradiated extremely important
Acute Effects of Whole Body Radiation

- Exposure interval (time), dose, fractionation, and dose rate critically important determining clinical endpoint
- Effects of whole body radiation significantly different compared to partial body or localized radiation
- “Classic” acute radiation syndromes (ARS) based on single fraction whole body exposure at high dose rates
- Syndromes follow three phases referred to as the prodromal phase, latent phase, and manifest illness
- Duration of each phase and interval between phases varies depending primarily on total dose and dose rate
- Mixed photon/neutron beams may worsen prognosis
Acute Effects of Whole Body Radiation

- Traditional ARS includes cerebrovascular syndrome, gastrointestinal syndrome, and hematopoietic syndrome.
- Recent approaches to the classification of ARS have shifted to five tiers of predicted clinical severity:
  - Mild: 1-2 Gy
  - Moderate: 2-4 Gy
  - Severe: 4-6 Gy
  - Very Severe: 6-8 Gy
  - Lethal: > 8 Gy
- Predicted onset of symptoms, clinical manifestations, and laboratory findings developed for each category.
- Overall prognosis and treatment recommendations provided for each of the five classifications.
- Refer to single page handout.
Radiation Induced  
Oncogenic Transformation

- Radiation capable of producing genetic changes  
- Genetic alterations shown to be the cause of cancer  
- Cancer development to two contributing processes  
- Conversion of proto-oncogenes to oncogenes represents the gain of oncogenic potential  
- Loss of tumor suppressor genes (emerogenes) represents the loss of anti-oncogenic potential  
- Emergence of radiation induced oncogenic phenotype secondary to “balance” of transformation & cell killing  
- Refer to single page handout
Thank you for your attention

- Questions
- Comments
- Discussion