ICRP –2

Radiation biology studies – from cell culture to epidemiology

Expectations for radiation biology are that it is possible to extrapolate low dose and low dose rate effects from high radiation dose studies – this may not be possible…

Epidemiological data are often contradictory, mouse data as well

- Many studies on mRNA and protein expression document distinctly different patterns of gene expression for moderate and low doses of radiation

Even cell culture studies confirm that this is an erroneous assumption:

- E.g. in stem cells both hypersensitivity and hormesis

Differences between High- and Low-Dose Radiation Responses

Low Dose < 0.2 Sv

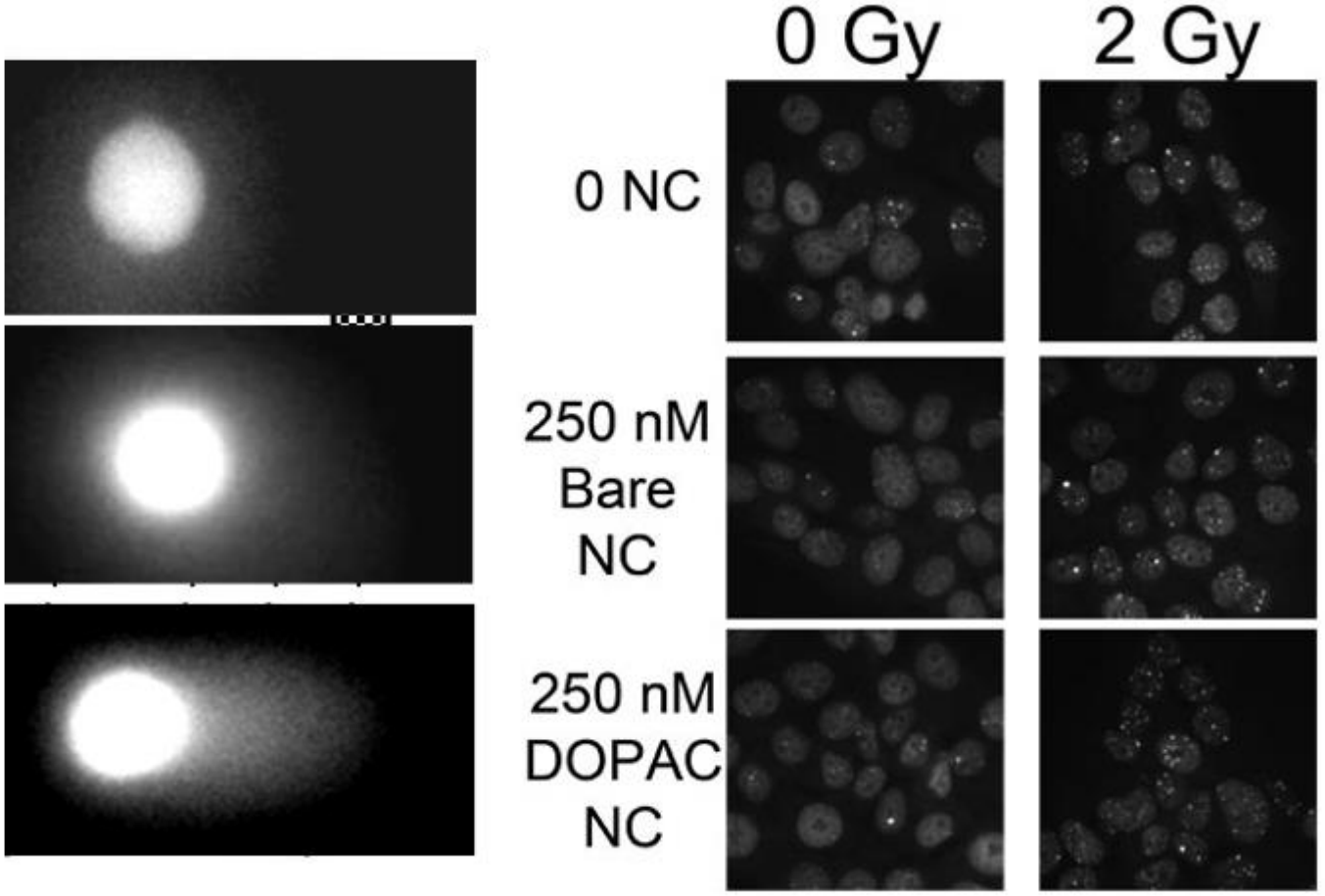
Cell killing low
DNA damage low/not detected
Gene Expression (Protective?)
Epigenetic Effects (Protective)
Free Radicals decreased
Indirect Action
MnSOD
Glutathione
↑ Selective Apoptosis
↓ Mutation Frequency
↓ Cell Transformation
Immune response? (+)
Cancer (??? %/mSv)?

High Dose > 0.2 Sv

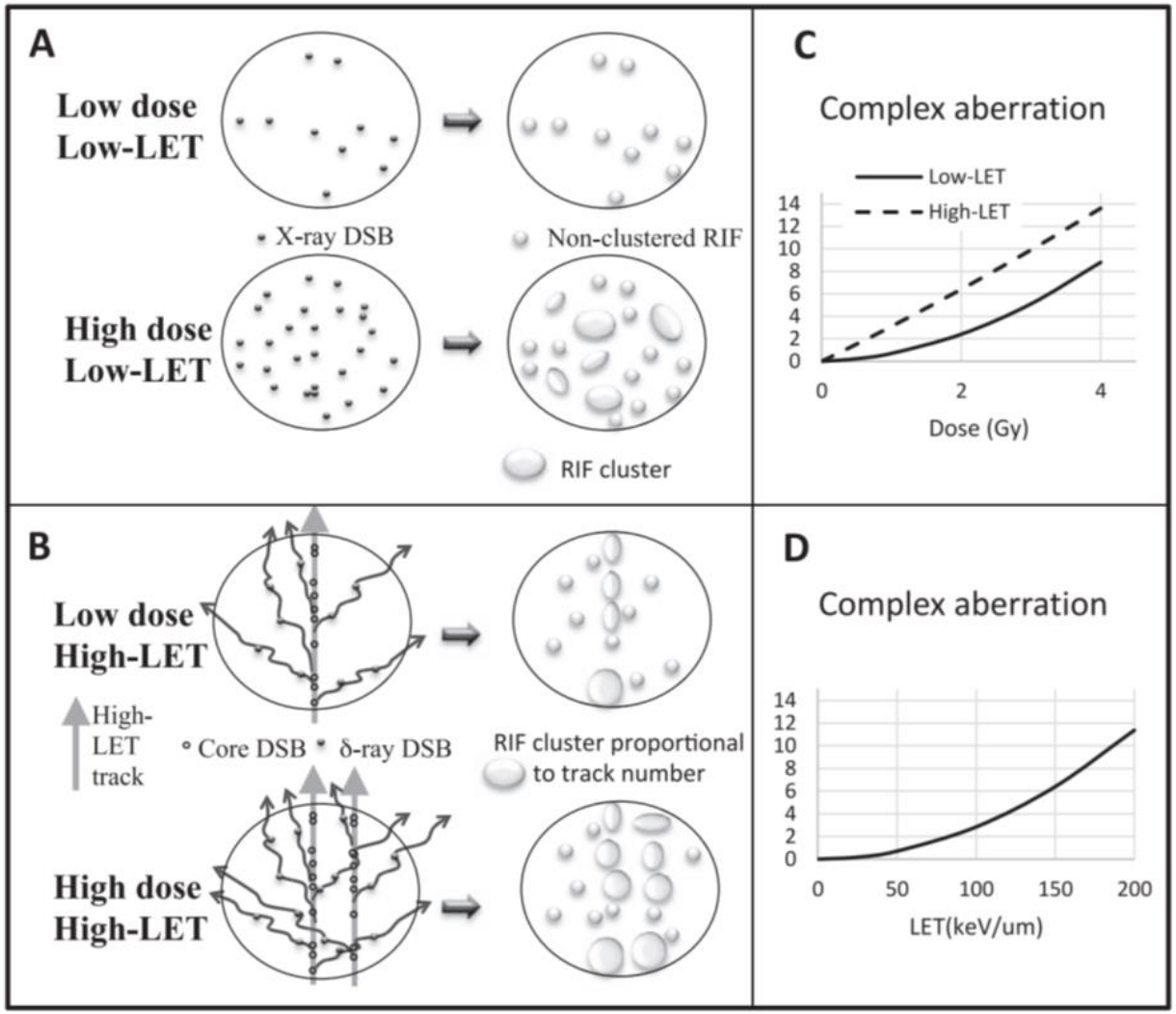
Cell killing high
DNA damage high
Gene Expression (Damage?)
Epigenetic Effects?
Free Radicals Increased
Direct Action

↑ Apoptosis (Increased)
↑ Mutation Frequency
↑ Cell Transformation
Immune response (-)
Cancer increased (5%/Sv)

Single cell assays for evaluation of DNA damage



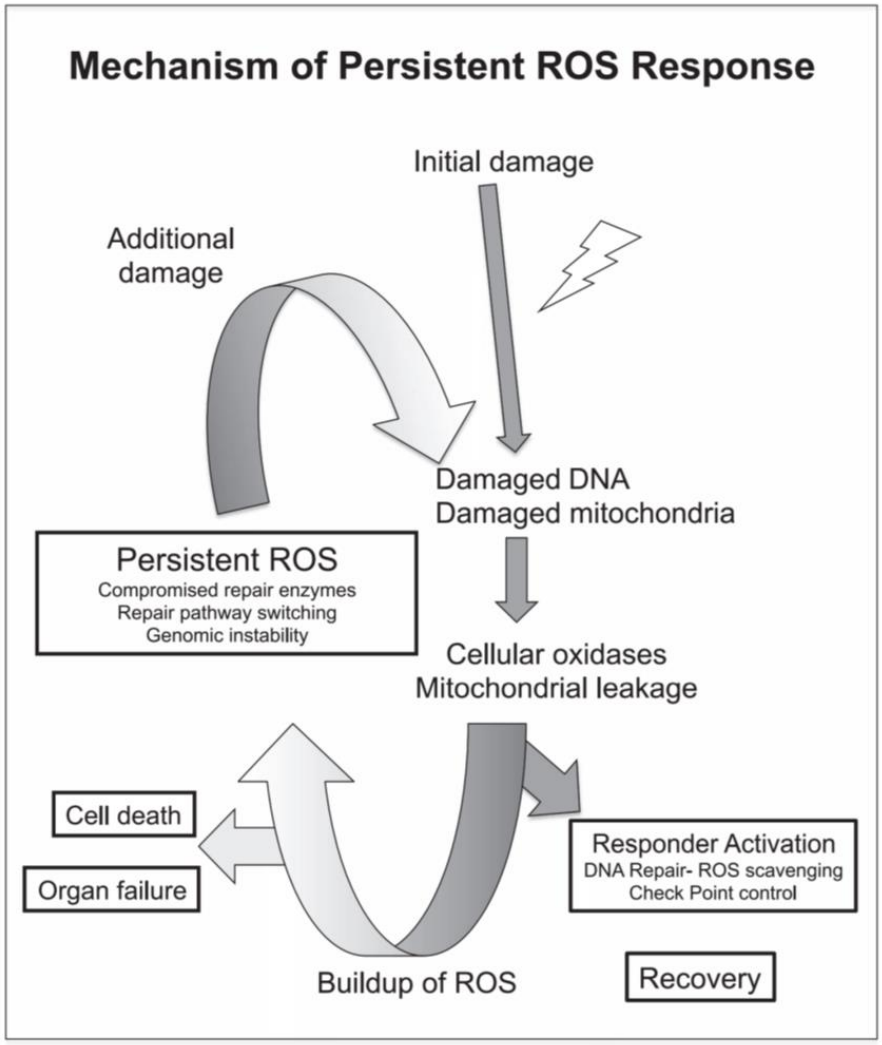
Radiation Induced Foci



Dose dependence of RIF clustering. Panel A: shows the formation of radiation-induced foci (see ref. 252) from double-strand breaks (DSB) in nuclei exposed to low and high doses of X ray. Panel A shows that RIF clustering would be expected to occur predominantly after high dose. Panel B: A similar representation for low and high doses of high-LET ions showing RIF clustering occurs along the track independent of the dose. Panels C and D: A consequence of clustering is the formation of complex chromosome aberrations, thus it would be expected that complex chromosome aberrations would differ based on dose (panel C) and LET (at a constant fluence) (panel D). Panels C and D: Y-axis are number of complex chromosome aberrations.

Sridharan, D. M., Asaithamby, A., Bailey, S. M., Costes, S., Doetsch, P. W., Dynan, W., Kronenberg, A., Rithidech, K. N., Saha, J., Snijders, A. M., Werner, E., Wiese, C., Cucinotta, F. A. and Pluth, J. M. Understanding Cancer Development Processes after HZE-Particle Exposure: Roles of ROS, DNA Damage Repair, and Inflammation. Radiat. Res. 183, 1–26 (2015).

DNA Damage Feedback Loop



Proposed positive-feedback loop between ROS and DNA damage. Initial interaction of radiation damages DNA and mitochondria, which stimulates formation and release of ROS. High levels of ROS lead to various responses, including an increase in ROS scavenging and DNA repair protein activities and activation of DNA damage-dependent cell cycle checkpoint controls. In some cases, this responder activation is sufficient to promote recovery, and in others, an overwhelming amount of damage may lead to cell death and organ failure. We suggest, however, that for high-LET radiation-induced damage, there may be conditions where cells neither recover nor die, but rather adopt a state of persistent oxidative stress, characterized by compromised DNA repair enzymes, reliance on less accurate mechanisms of repair and genome instability. These effects lead via signaling mechanisms to additional ROS release, perpetuating the cycle.

Sridharan, D. M., Asaithamby, A., Bailey, S. M., Costes, S., Doetsch, P. W., Dynan, W., Kronenberg, A., Rithidech, K. N., Saha, J., Snijders, A. M., Werner, E., Wiese, C., Cucinotta, F. A. and Pluth, J. M. Understanding Cancer Development Processes after HZE-Particle Exposure: Roles of ROS, DNA Damage Repair, and Inflammation. Radiat. Res. 183, 1–26 (2015).

Single cell assays for evaluation of DNA damage

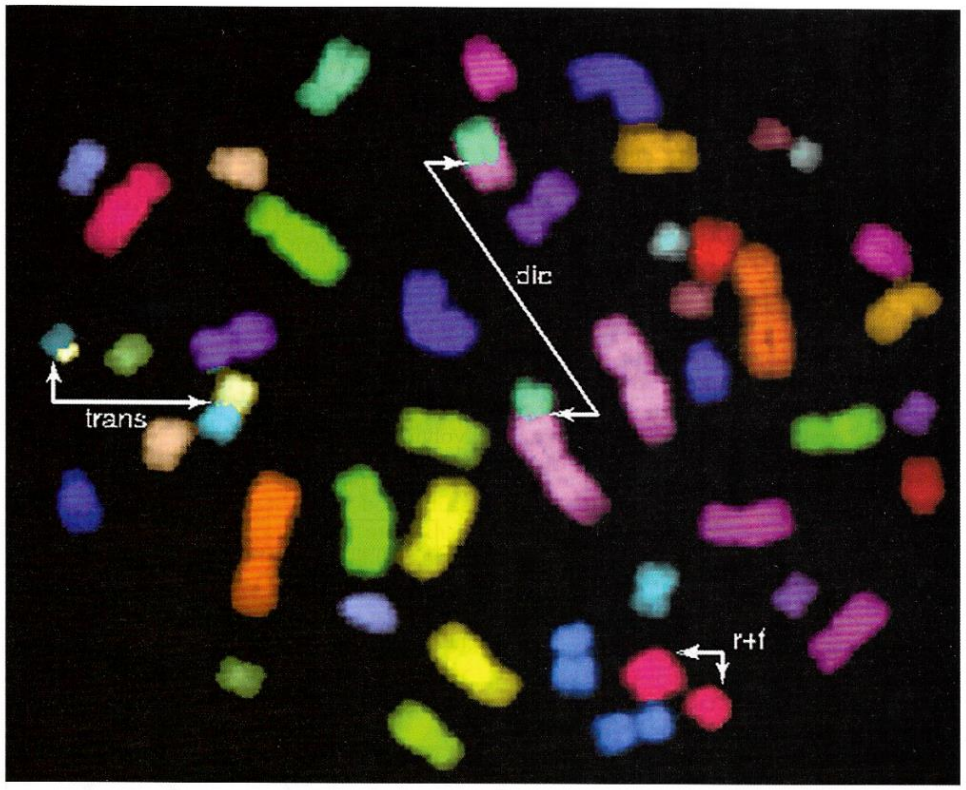


FIGURE 2.18 Fluorescence *in situ* hybridization of a metaphase spread from a cell that received 4 Gy. The hybridization was performed with a cocktail of DNA probes that specifically recognize each chromosome pair. Chromosome aberrations are demarcated by the *arrows*. (Courtesy of Dr. Michael Cornforth.)

E. J. Hall and A. J. Giaccia, "Radiobiology for the Radiologist," 8th Edition, Lippincott Williams & Wilkins, Philadelphia, 2018. pg. 31 Fig. 2.18

Cell population assays for evaluation of cell death

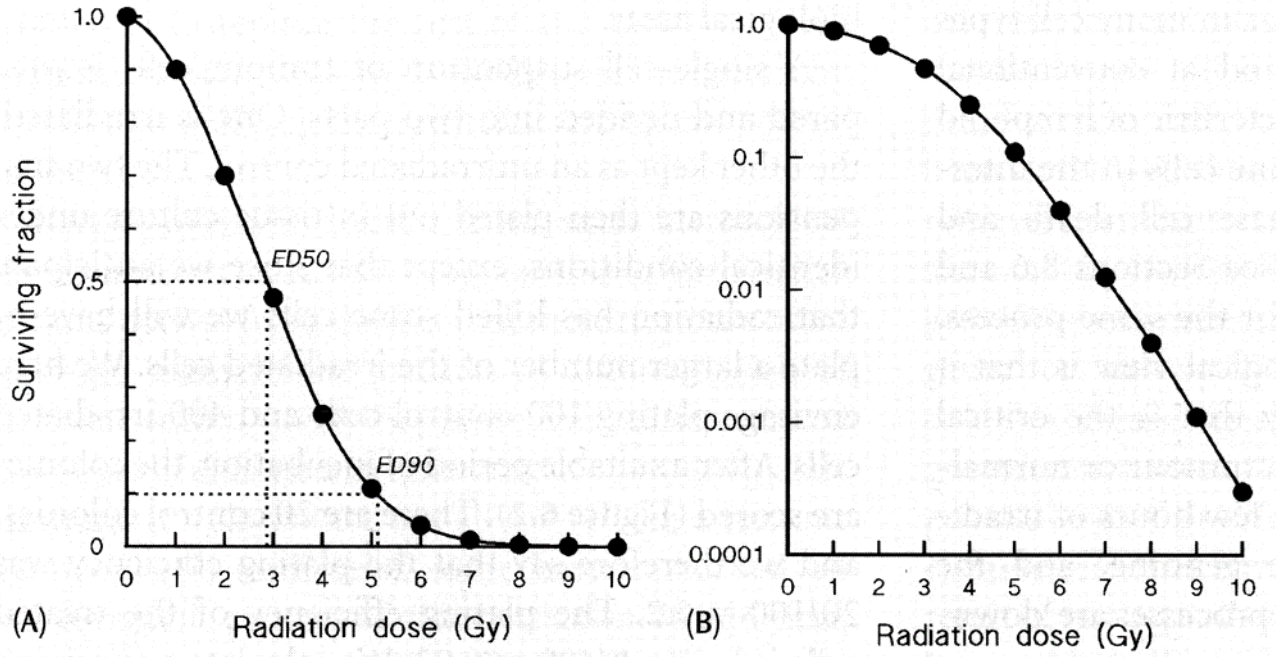


Figure 6.3 *A typical cell survival curve for cells irradiated in tissue culture, plotted (A) on a linear survival scale. (B) Shows the same data plotted on a logarithmic scale.*

Table 2 – Signalling molecules involved in producing bystander effects.

Intracellular signalling molecules

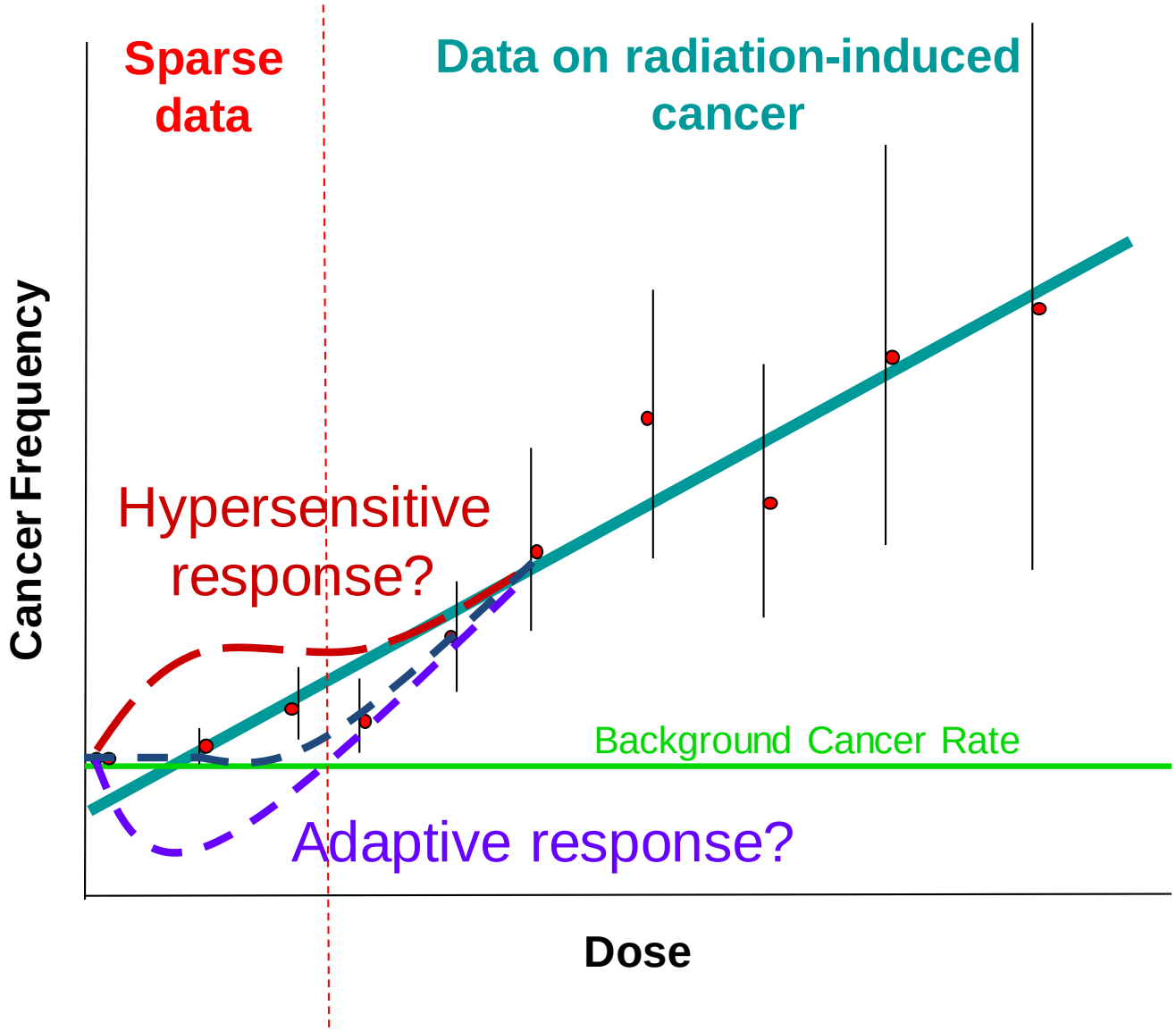
- (p53) Tumour protein 53
- (CDKN1A, p21) Cyclin-dependent kinase inhibitor 1A
- (MAPK) Mitogen-activated protein kinases
- (ATR) Ataxia telangiectasis and Rad3 related
- (DNA-PK) DNA-dependent protein kinase
- (PKC) Protein kinase C
- (ATM) Ataxia telangiectasia mutated protein

Intercellular signalling molecules

- (ROS) Reactive oxygen species
- (NO) Nitric oxide
- (5-HT serotonin) 5-hydroxytryptamine
- L-DOPA
- Glycine
- Nicotine
- Interleukin 8
- (RNS) Reactive nitrogen species
- (TGFβ1) Cytokines
- TNFα

Marín et al., Bystander effects and radiotherapy. Reports of Practical Oncology & Radiotherapy, Volume 20, Issue 1, January–February 2015, Pages 12–21

Linear Non-Threshold Radiation Dose-Response Model – Good for High Doses



Dose and Dose Rate Effectiveness Factor (DDREF)

Definition

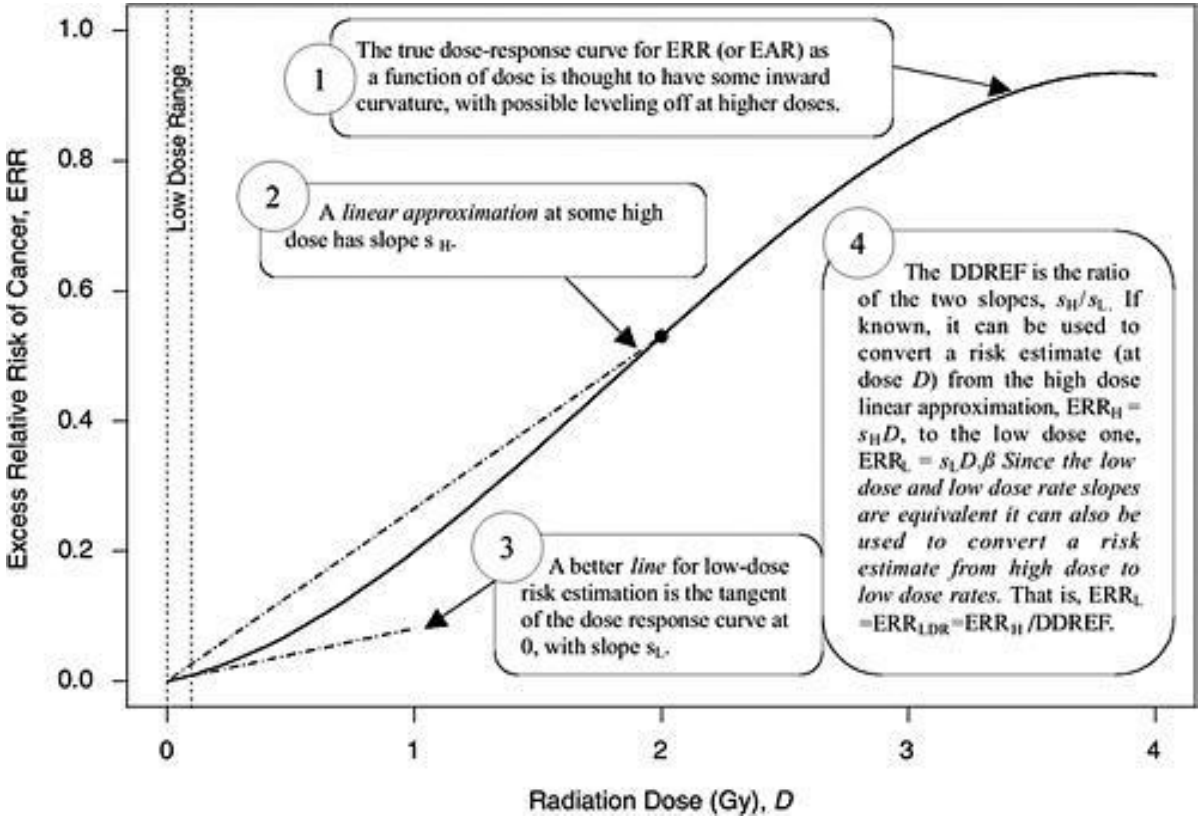
The risk observed from acute exposures is divided by DDREF in order to determine the risk of protracted exposure.

Example

If a 1 Gy acute exposure increases cancer risk by 10% and DDREF is 2, then a 1 Gy exposure spread over a year will increase cancer risk by 5%.

- **Sources of Information for DDREF Estimate**
- LSS Cohort (A-bomb survivors data)
- Animal studies

Dose and Dose Rate Effectiveness Factor (DDREF)



ERR

- $\alpha * Dose + \beta * Dose^2$

DDREF

- acute / protracted
  $(\alpha * D + \beta * D^2) / (\alpha * D)$
  $= 1 + \beta/\alpha * Dose$

LSS DDREF

(at 1 Gy)

- Estimated to be 1.5 (1.1 - 2.3) by the BEIR VII committee.

- FIGURE 10-1 A hypothetical dose-response curve with a linear approximation for low doses (*i.e.*, the tangent of the curve at dose zero) and a linear approximation based on risk at one particular high dose (*i.e.*, the line that passes through the origin and the true dose-response curve at the high dose), when the high dose is taken to be 2 Gy. The DDREF at this high dose is the larger slope divided by the smaller slope.
  Beir VII Phase II Figure 10-4

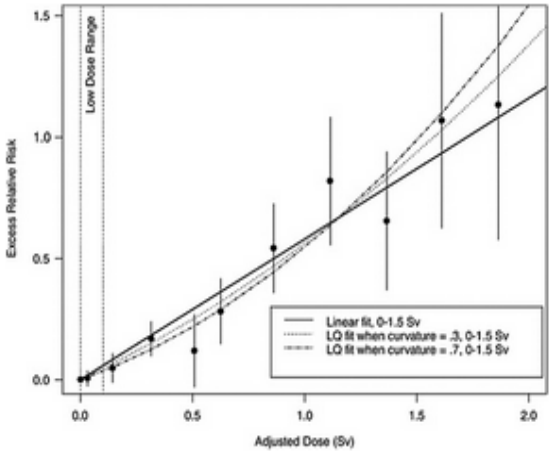
DDREF estimate per BEIR VII

Seventh report of the Biological Effects of Ionizing Radiation (BEIR) committee estimates a 3-12% absolute increase in the risk of fatal cancer development per Sievert (Sv) of exposure (National Research Council, 2006).

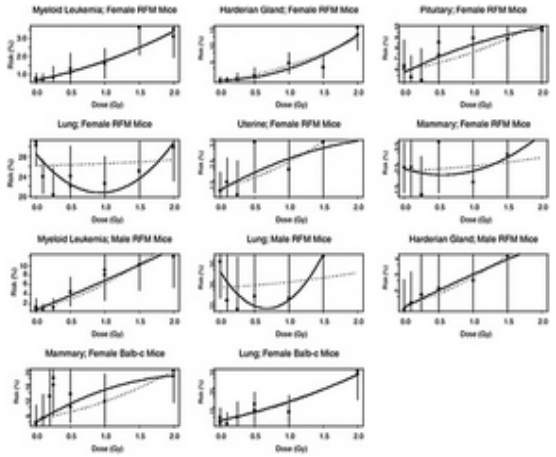
The BEIR VII committee used

- atomic bomb survivor data to evaluate dose and dose rate effectiveness factor for the life span study of atomic bomb survivors (DDREF);
- dose-response data from a series of large mouse studies carried out at the Oak Ridge National Laboratory in the late 1970s involving whole body gamma exposures from a cesium-137 source

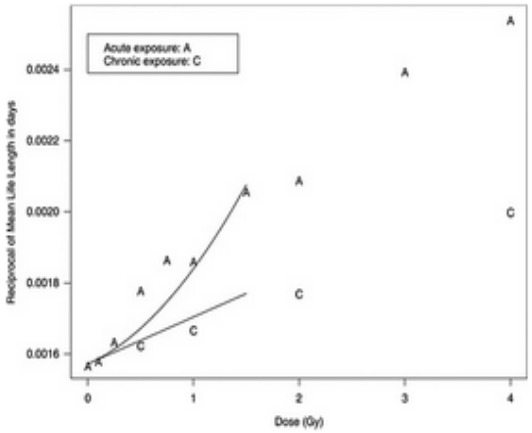
DDREF estimate per BEIR VII



LSS carcinogenesis
DDREF ~ 1.3 (0.8, 2.4)



Animal carcinogenesis
DDREF ~ 1.4 (1.1, 2.6)



Animal mortality
DDREF ~ 2.0 (1.3, 7.7)

Figures 10-2, 10B-2, and 10B-3 of "Health risks from exposure to low levels of ionizing radiation: BEIR VII phase 2" . **Linear quadratic models used for DDREF evaluation fit to three data sources, excess risk of carcinogenesis in atomic bomb survivors (LSS carcinogenesis), risk of tumor development in various animal studies (Animal carcinogenesis), and inverse mean lifespan in two animal studies (Animal mortality).** DDREFLSS estimates derived from each data source are shown with 95% confidence intervals in parenthesis. These estimates were combined to form BEIR VII's central estimate, DDREFLSS ~ 1.5 (1.1, 2.3).

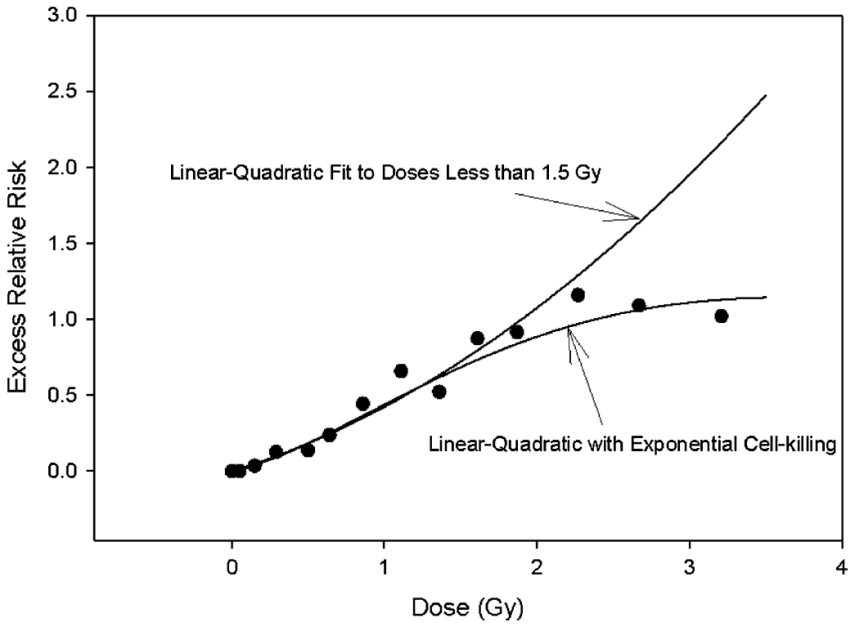
Haley et al in preparation.

DDREF estimates depend on dose limits chosen

Table 1. DDREF estimates as a function of data used.

	Maximum dose included in the analysis (Gy)							
	1.0	1.25	1.5	1.75	2.0	2.5	3.0	All
LSS DDREF	1.37	1.93	1.39	1.38	1.34	1.27	1.15	1.11
UNSCEAR DDREF	1.52	2.03	1.36	1.34	1.28	1.18	1.09	1.07

The dependence of the dose restriction value in the BEIR VII method with the UNSCEAR DDREF definition is calculated at 1 Gy



Fitted dose-response functions to the LSS solid cancer incidence data: a) linear-quadratic using data less than 1.5 Gy exposure as in BEIR VII; b) linear-quadratic with an exponential cell-killing term using the complete LSS incidence data set.

Hoel, Health Physics 2015 108(3)

DDREF estimates from abbreviated animal life shortening data

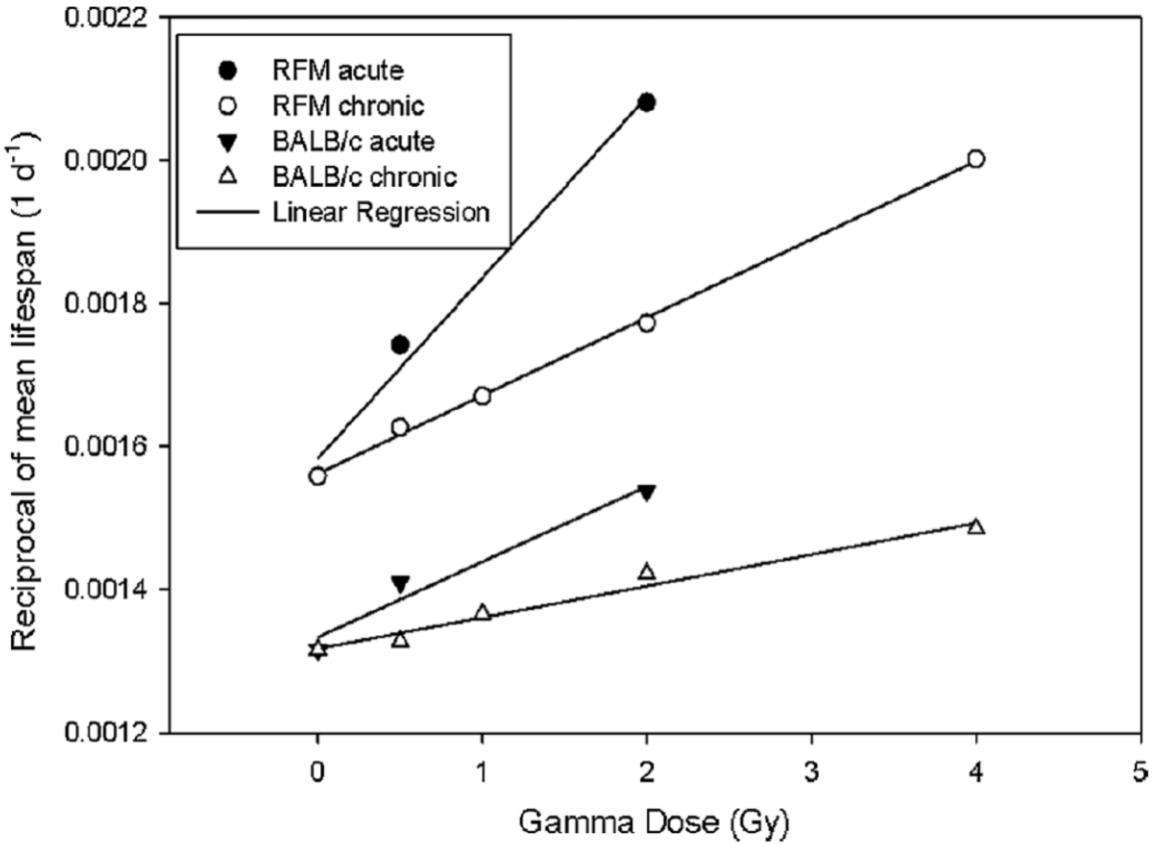
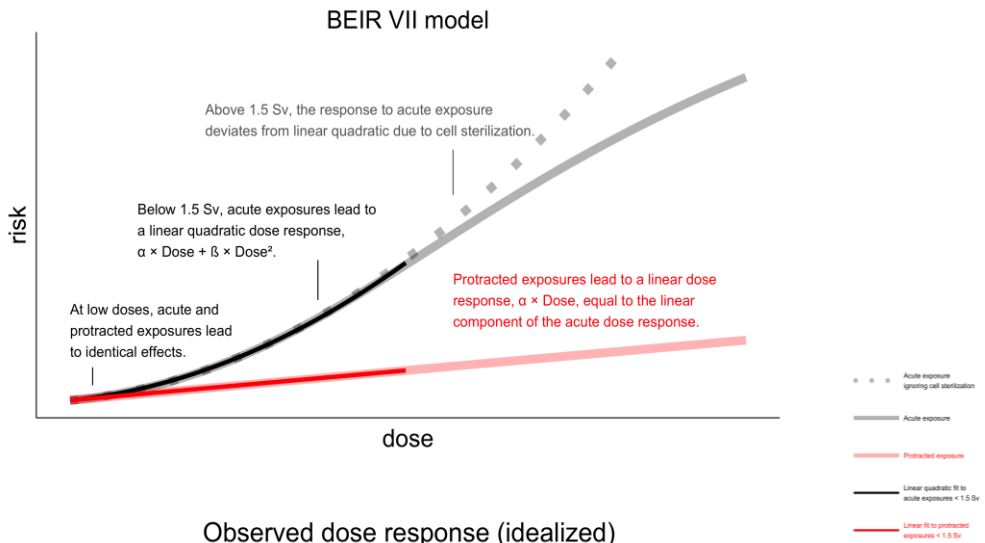


Fig. 4. Linear fits to the reciprocal mean lifetimes of the gamma exposed RFM and BALB/c female mice. Both acute and chronic exposures are shown.

Hoel, Health Physics 2015 108(3)

Replicating DDREF evaluation using more of the existing animal data



A schematic representation of the BEIR VII dose response model (top) is shown above an idealized representation of observations (bottom). Each panel shows dose (x-axis) vs. risk (y-axis) where risk represents the excess risk of carcinogenesis or organism mortality.

Black lines represent the response to acute exposures.

Red lines represent the response to protracted exposures.

Thick semi-transparent lines show the dose response curve implied by the BEIR VII model. Dotted transparent lines in the BEIR VII model represent a hypothetical acute dose response if no cell reproductive death occurred.

Thin opaque lines show the expected fit of a linear quadratic model to exposures less than 1.5 Sv in the BEIR VII model (top) vs. observations (bottom).

Haley et al in preparation.

NURA and JANUS

- Northwestern University Radiation Tissue Archives (NURA)
  - paraffin embedded dog, mouse and rat tissue samples from ANL (JANUS experiments) as well as ITRI, PNNL and UCDavies
  - two websites:
  - http://janus.northwestern.edu/dog_tissues/introduction.php
  - http://janus.northwestern.edu/janus2/index.php

γ|n⁰ Janus Tissue Archive

lookup animal by id 1.1

Dosimetry

	Min	Max
Total Dose (cGy)	0	3664
Dose Rate (cGy/min)	0	38
Fractions	0	1000

Radiation type: All (•) Control ( ) Gamma ( ) Neutron ( )

Demography

	Min	Max
Age at death (days)	134	2367
Age at first treatment	17	1400

Gender: Either (•) Male ( ) Female ( )

Micro Pathologies

Macro Pathologies

Janus Experiments

search reset

Introduction

This web application enables interested parties to search autopsy records from the mice used in the Janus Irradiation Experiments and request histological samples from animals of interest.

Created by Dave Paunesku for the Woloschak Lab at Northwestern University and financed by NASA and the US Department of Energy. To report problems or make suggestions, please contact Ben Haley.

Instructions

Use the boxes to the left to search for specific animals. Click the question mark image for help regarding the corresponding search criteria or the double arrow to expand closed search boxes.

Registration

If you would like to create an account to upload slide images from these animals, please contact Dr Tatjana Paunesku. We will be happy to provide you with the necessary credentials.

γ Beagle Dog Tissue Archive

Search for dog data and tissues.

Dog # Lookup

Advanced Search

Dog Details

Organs & Tissues

Only Include Results with Tissues

Experiments

All Experiments

- Cobalt Chronic Exposures (1167)
- Cobalt Acute and Fractionated Exposures (128)
- Controls (175)

search reset

This archive contains data and specimens from dogs irradiated by Thomas Fritz, William Norris, and Tom Seed at Argonne National Laboratories between 1954 and 1991. Samples from our tissue bank are available on request. For irradiated mouse tissues please visit the Janus Tissue Archive.

Brought to you by NASA and the Department of Energy. Thank you to Angela Babbo, Tom Fritz, Christine Gerin, Dave Paunesku, Tanja Paunesku, Beau Wanzer, Charles Watson, Gayle Woloschak, and the rest of Wololab.

To report problems contact Ben Haley.

ANL: external beam dog studies

Using NURA mouse tissue samples (JANUS archive mostly at the moment)

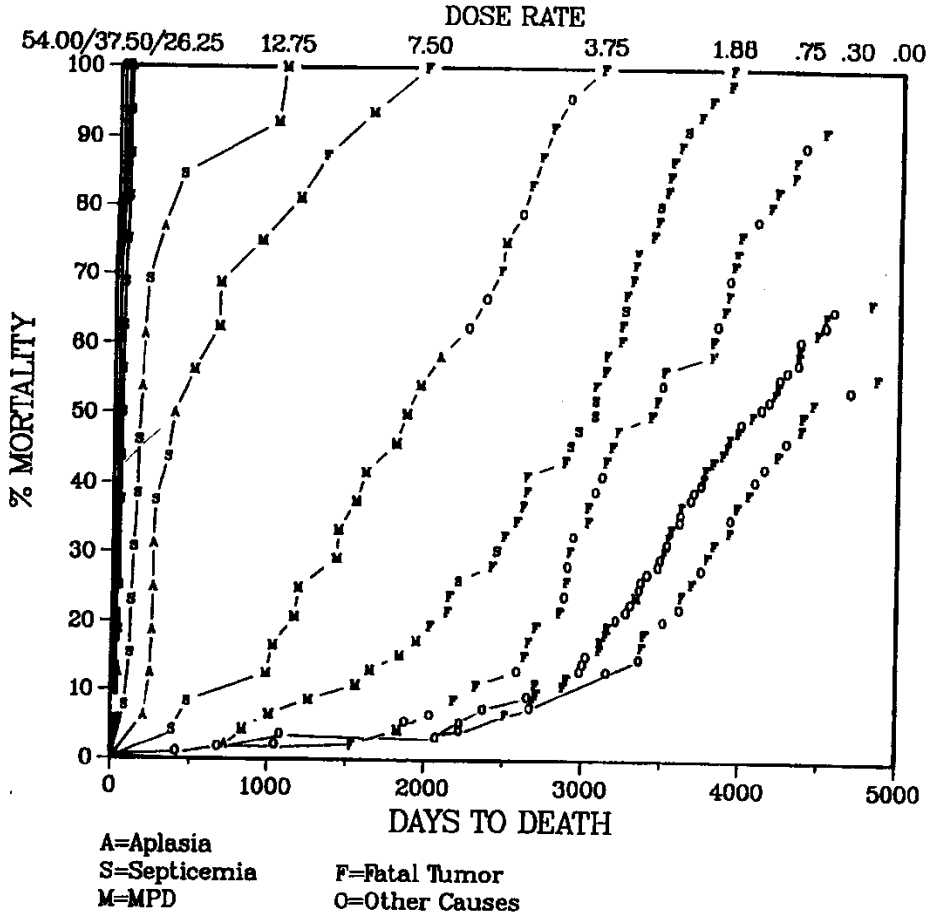
NURA and other data sources (digitized, archived, centralized, crosschecked against published literature) such as the International Radiobiology Archive (IRA) and European Radiobiology Archive (ERA) can be used for different types of statistical analysis.

NURA samples can be used for:

- mitochondrial DNA evaluation
- micro RNA studies
- histological examination
- elemental distribution in tissues (XFM)

A typical research project using NURA includes researching the data archive, selecting the tissues to be sectioned and processing them for regular histopathology, high throughput X-ray fluorescence elemental microscopy, or subjecting them to a variety of molecular analysis techniques focusing on proteins, DNA or micro RNAs.

Dog data analyses were limited



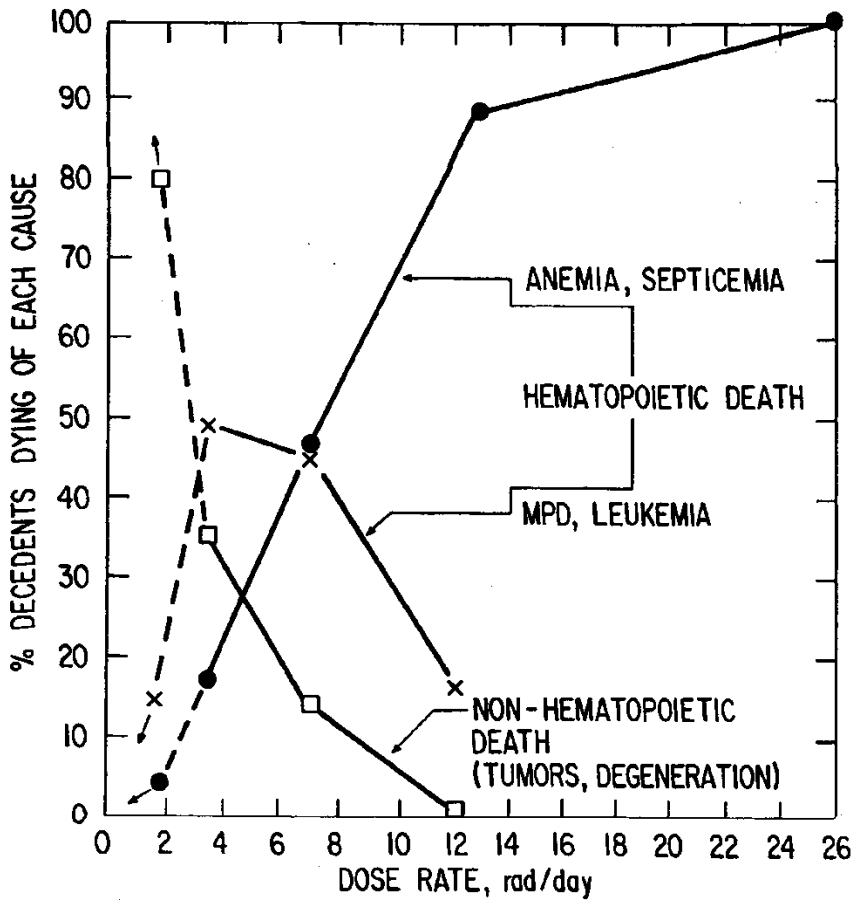


Figure 2. Relationship between dose rate, time to death and causes of death in adult beagles continuously irradiated (22 h day^{-1}) at several dose rates (rads day^{-1}; average absorbed dose).

Fritz T. (2002) BJR Supplement 26, 103-111.

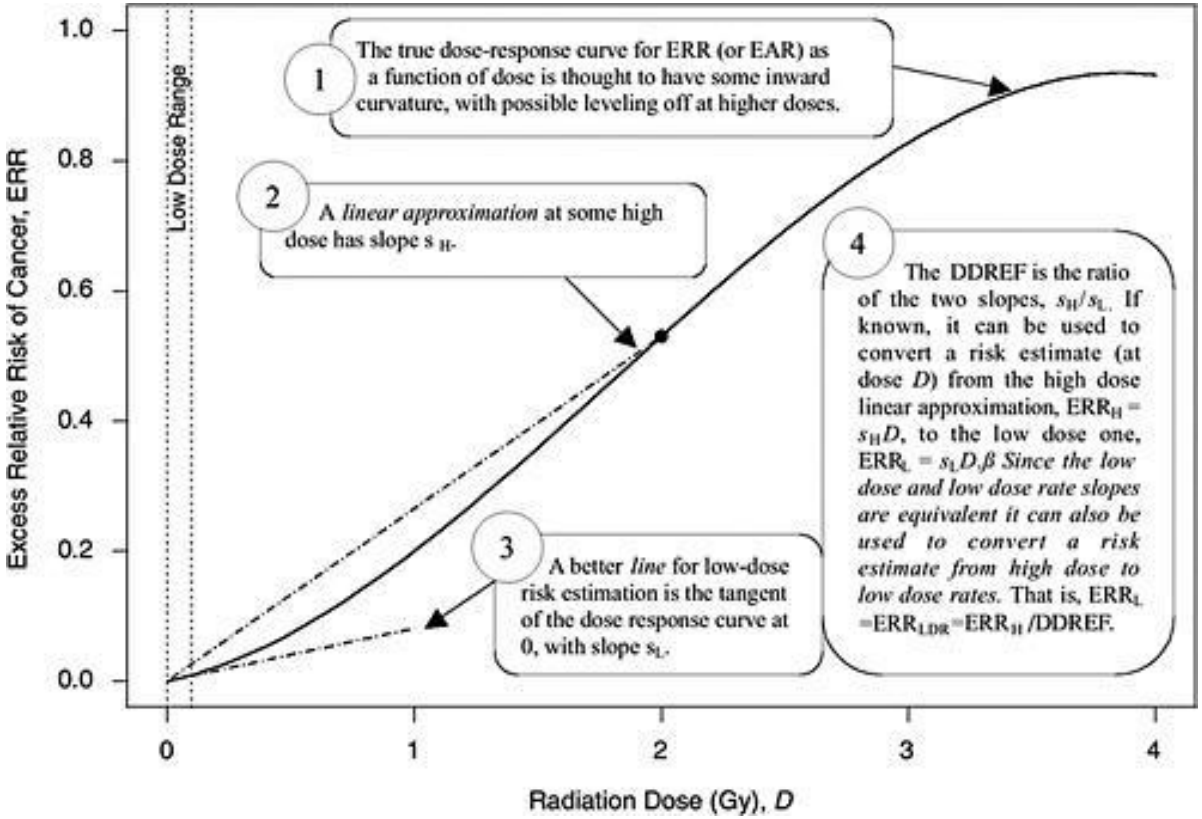
Figure 1. Relationship between causes of death and daily dose rate (average absorbed dose) in adult beagles exposed continuously (22 h day ^{1}) to ^{60}Co γ-irradiation.

Fritz T. (2002) BJR Supplement 26, 103-111.

DDREF estimates – possible future

- A new dose response model is needed to develop an estimate that is not biased by arbitrary factors in the data analysis.
- Animal data can be used to validate robustness of any such new model.
- Addition of data obtained on species other than mice may improve DDREF estimates
- Preliminary DDREF re-estimates suggest that separation of dose effects and dose rate effects is needed

Dose and Dose Rate Effectiveness Factor (DDREF)



ERR

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LSS DDREF

(at 1 Gy)

- Estimated to be 1.5 (1.1 - 2.3) by the BEIR VII committee.

- FIGURE 10-1 A hypothetical dose-response curve with a linear approximation for low doses (*i.e.*, the tangent of the curve at dose zero) and a linear approximation based on risk at one particular high dose (*i.e.*, the line that passes through the origin and the true dose-response curve at the high dose), when the high dose is taken to be 2 Gy. The DDREF at this high dose is the larger slope divided by the smaller slope.
  Beir VII Phase II Figure 10-4

Ramifications of Low Dose Radiation Leads to Uncertainty

- Public's concern about radiation is increasing with every new accident and international test nuclear detonation, leading to:
  - Fear & "not in my backyard" attitudes (yet nuclear power plants are a green source of energy)
  - disproportionate cleaning expectations
  - Avoidance of medical diagnostic procedures
  - Misunderstanding among physicians about diagnostic exposures
  - Stress, fear, distrust of policies

Big data approaches allow follow-up of modest changes in gene expressions and enzyme activities

- Gene expression changes
  - microarrays
  - next-gen sequencing
  - ...
- Epigenetic changes
  - micro RNA
  - histone modifications
  - ...
- OMICS approaches
  - metabolomics
  - metalomics
  - ...