Factors which influence cellular radiosensitivity

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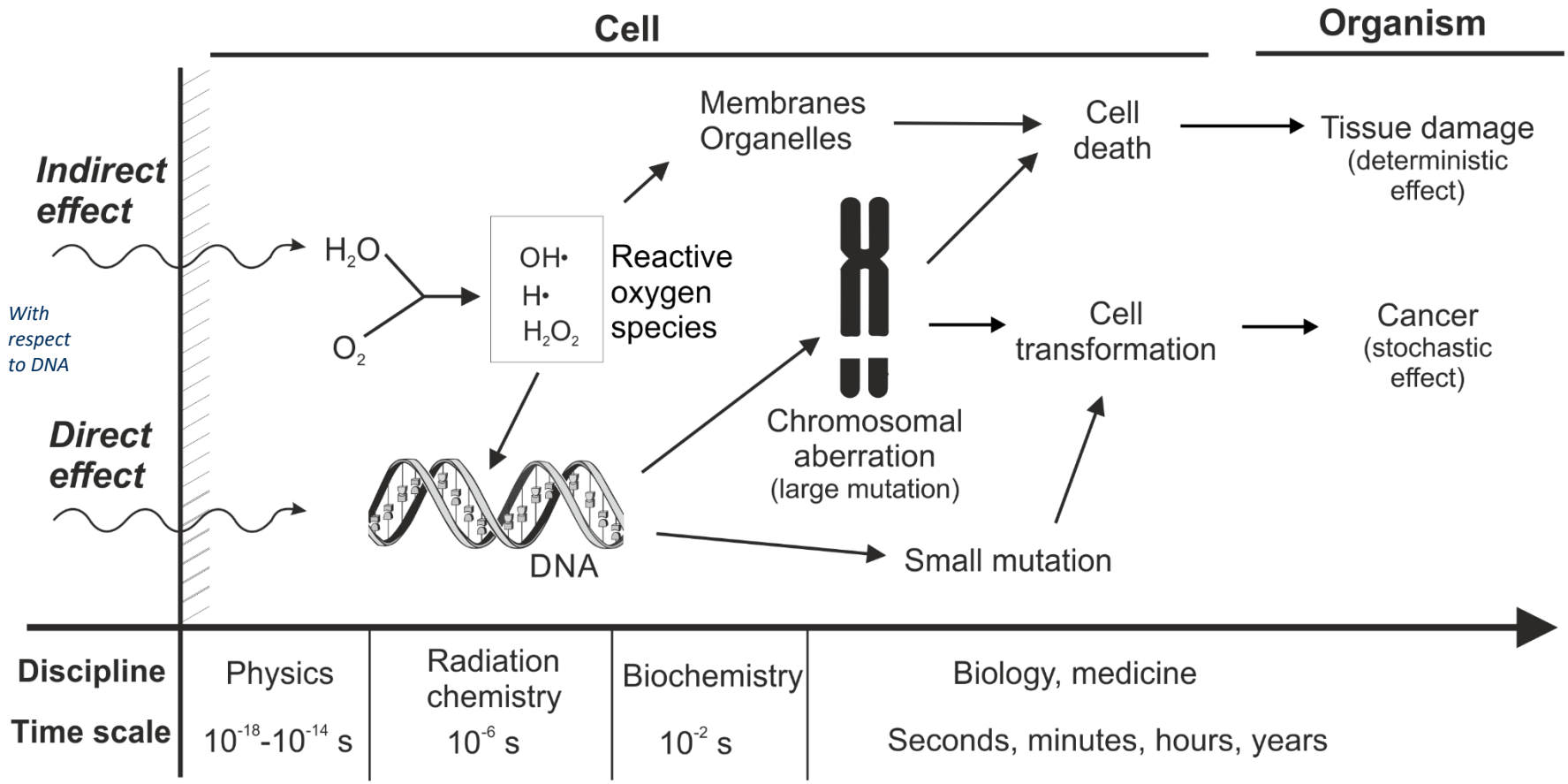
Events occurring in a cell exposed to ionising radiation



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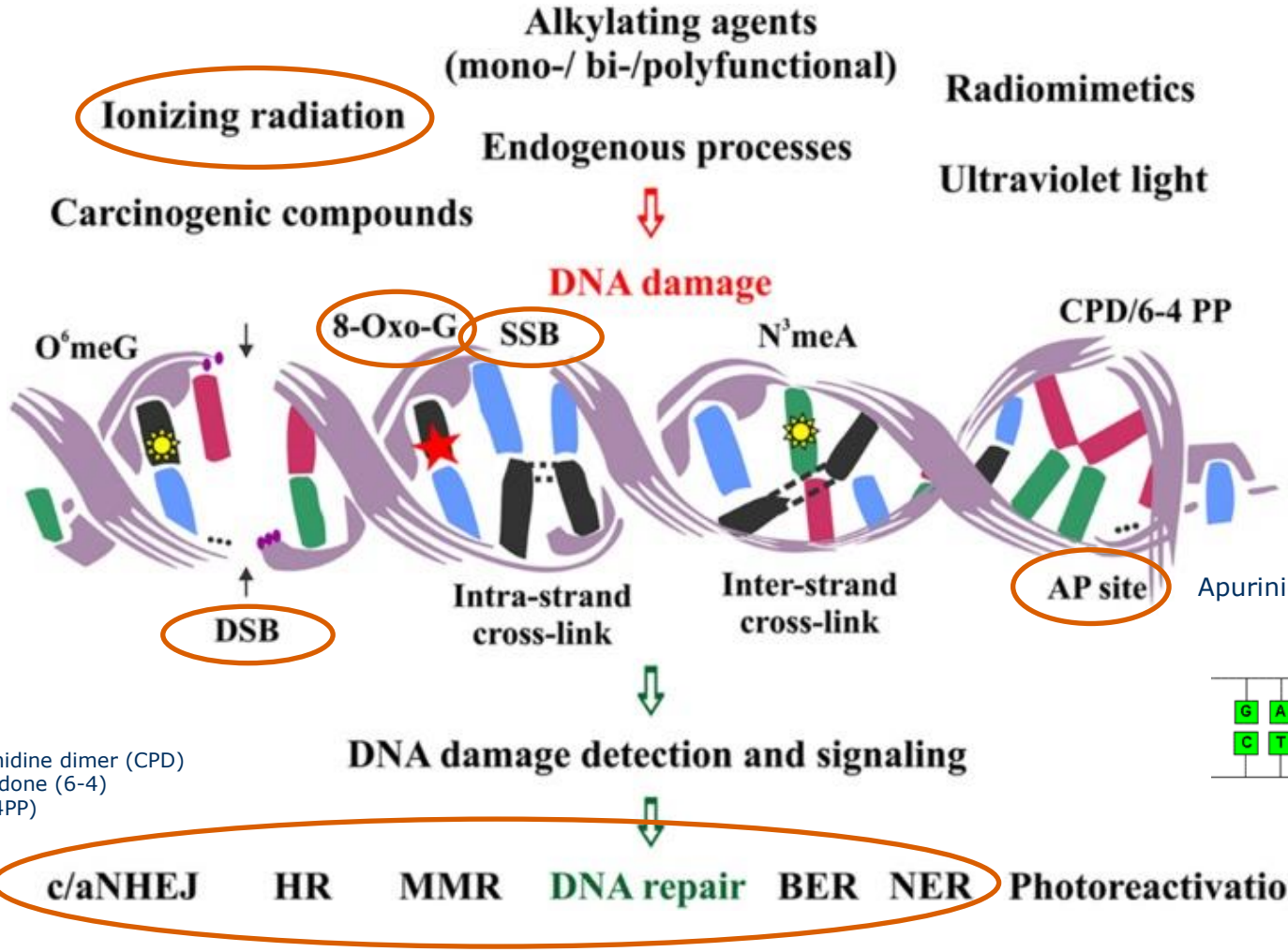
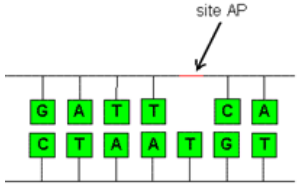
Lundholm L et al. EANM Technologist Guide 2016, chapter 4, www.eanm.org

DNA damage induction by radiation is partly overlapping that from other inducers



Names in full:
O^6-methylguanine
8-oxoguanine
N^3-methyladenine
cyclobutane pyrimidine dimer (CPD)
pyrimidine-pyrimidone (6-4)
photoproduct (6-4PP)

Apurinic/apyrimidinic site



Ionising radiation mainly induces double strand breaks (DSBs), single strand breaks (SSBs) and base damage (BD, such as 8-oxo-G, AP sites)

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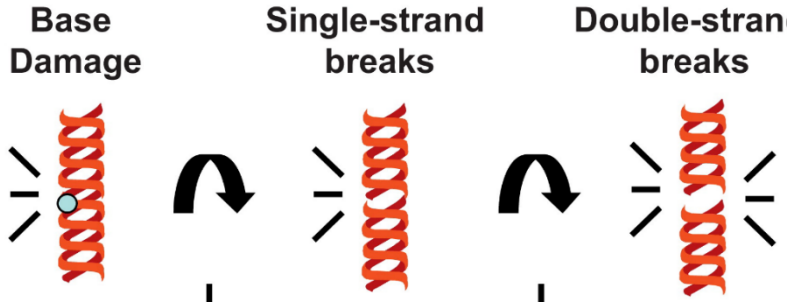
Manova et al. Front Plant Sci 2015

The cell needs time to repair DNA double strand breaks



Energy Microdeposition required	>1 eV/nm^3	>10 eV/nm^3	>100 eV/nm^3
Incidence per Gy per human cell	~ 10000	~ 1000	~ 40
50% repaired in:	5-10 min	10-20 min	> 50 min

Repaired by :

- **End-Joining** — Non-homologous end joining (NHEJ)
- **Recombination** — Homologous recombination (HR)
- **Excision-Resynthesis** — Base excision repair (BER), Nucleotide excision repair (NER), Mismatch repair (MMR)

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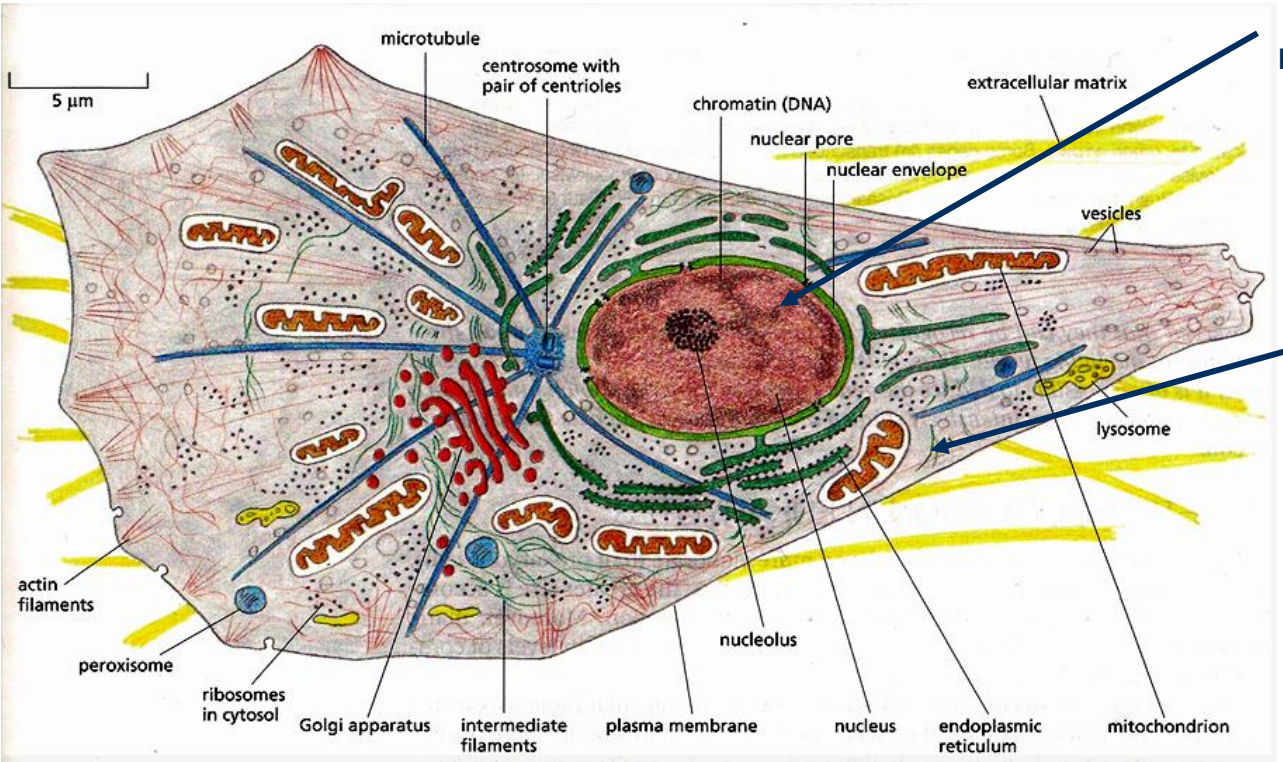
Lavelle and Foray. Int J Biochem Cell Biol. 2014

The cell nucleus is the main intracellular radiation target

Cells may:

- Survive an exposure without any detriment (due to efficient repair mechanisms)
- Survive after misrepair which may influence its function or the function of its descendants
- Die



The most sensitive cellular target for the action of ionising radiation (IR) is the cell nucleus which contains the DNA

The cytoplasm may be the main target for radiation in the low dose range and for bystander effects

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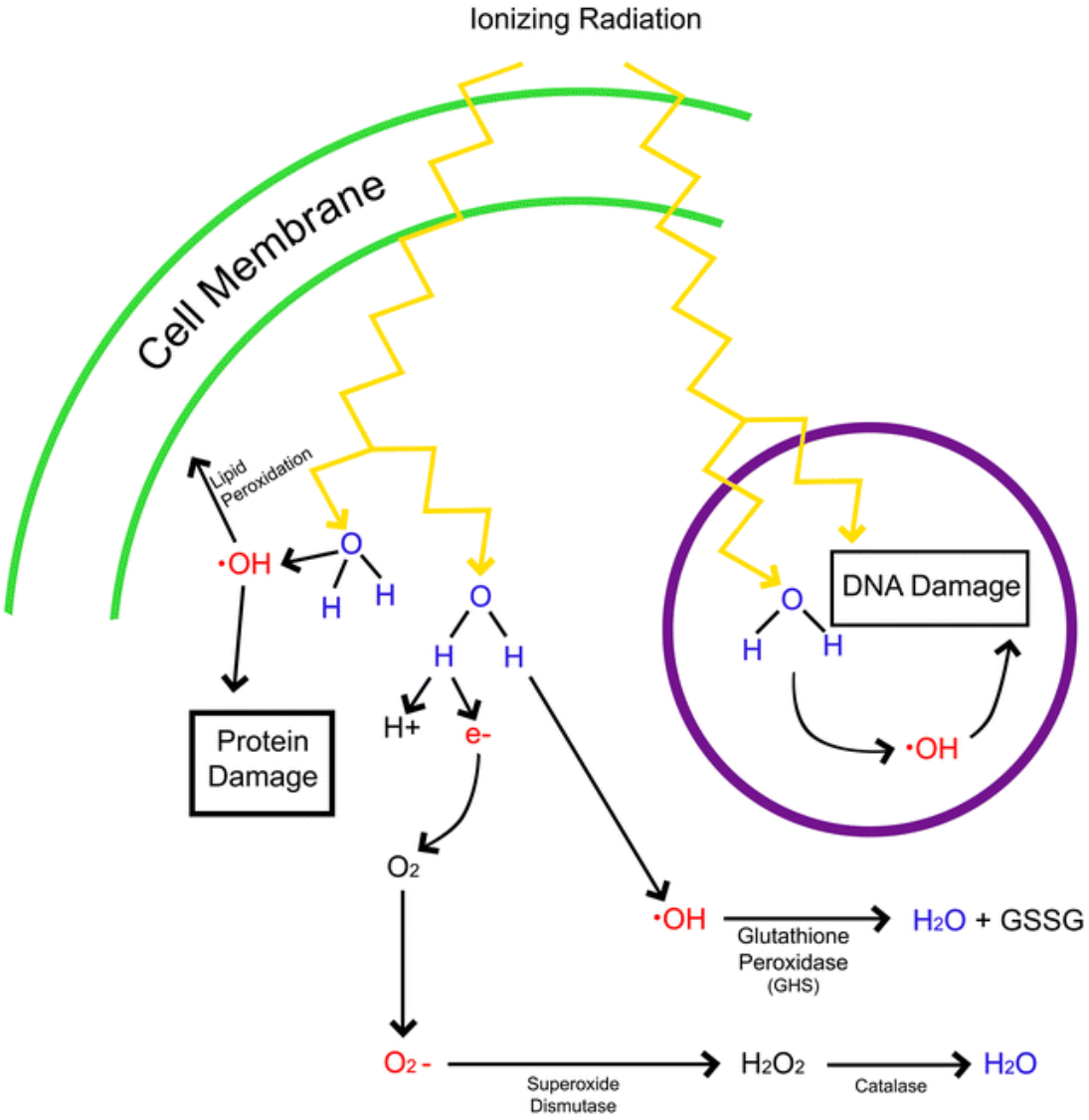
Effects on other organelles?



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IR can damage cellular proteins and lipids at very high doses

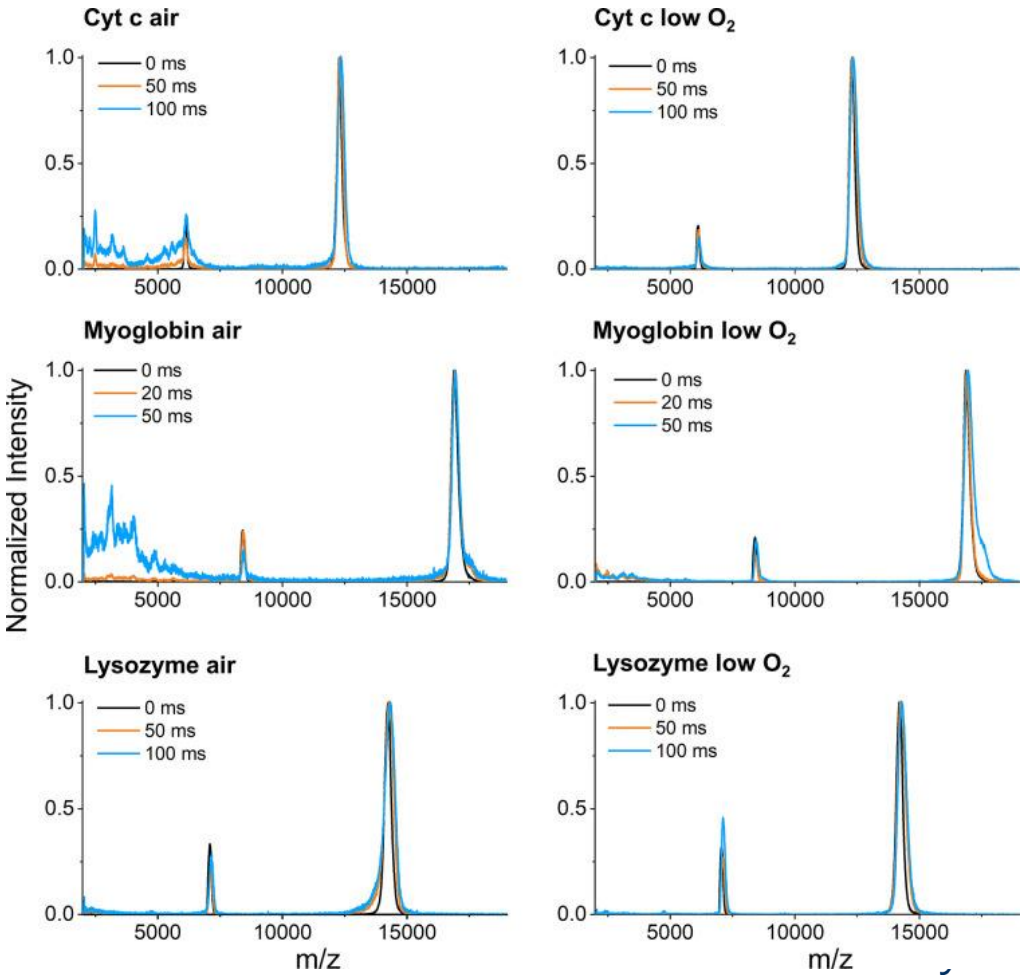
Proteins

- Ionizing radiation cause fragmentation and aggregation of protein molecules (Kumta, Nature, 1961)
- Radiation damage is a limiting factor in obtaining high-resolution structures in crystallographic experiments at synchrotrons (Nass, Acta Cryst, 2019)
- Mediators are in particular hydroxyl radicals (•OH)
- Importantly – doses needed are extremely high
  - "Doses as low as 100 Gy have been shown to cause radiation damage in the form of disulfide bond breakage"

Lipids

- Membranes

MALDI results show fewer lower MW products forming for cyt *c* and myoglobin sample preparations in low oxygen versus air-saturated solutions after X-ray exposure

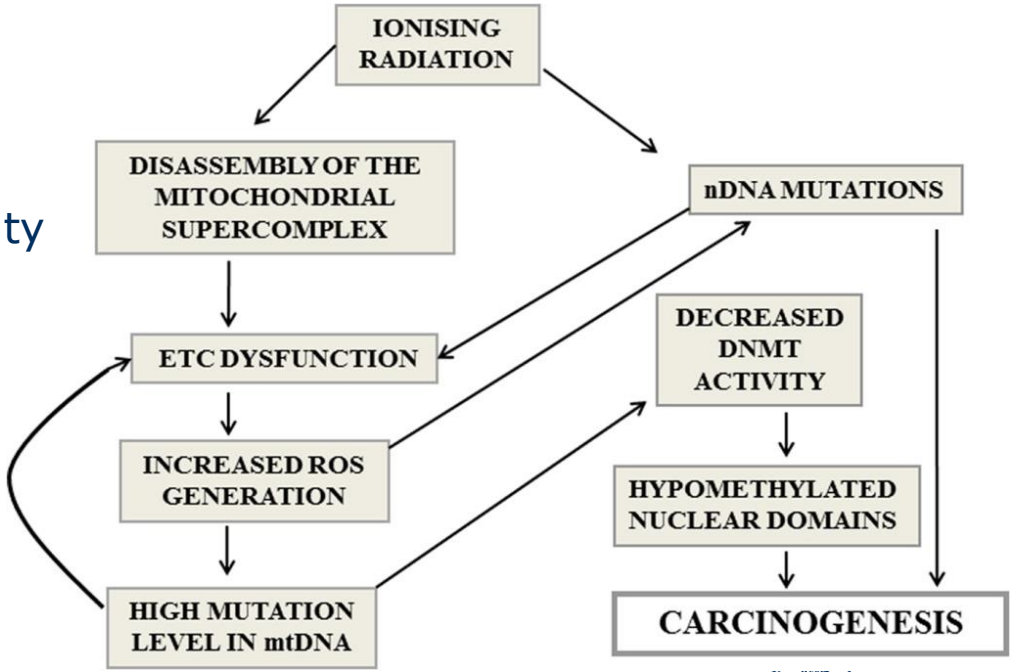


Kristensen et al. J Synchrotron Radiat. 2021

IR may modulate the epigenome and nuclear DNA via effects on mitochondria and reactive oxygen species

Mitochondria

- Oxidative stress by IR -> altered function of the electron transport chain - > persistent mitochondrial superoxide (O2-) production -> mtDNA mutation induction
  - High gene density
  - No shielding histones
  - ROS produced in close vicinity



Is epigenetic reprogramming responsible for the change in cell state, or is it a consequence of the change in cell state?

Zielske J Cell Biochem 2015

- Global DNA hypomethylation in normal cells (24 h after IR)

ETC; Electron transport chain
DNMT; DNA methyl transferase, which adds methyl groups on DNA

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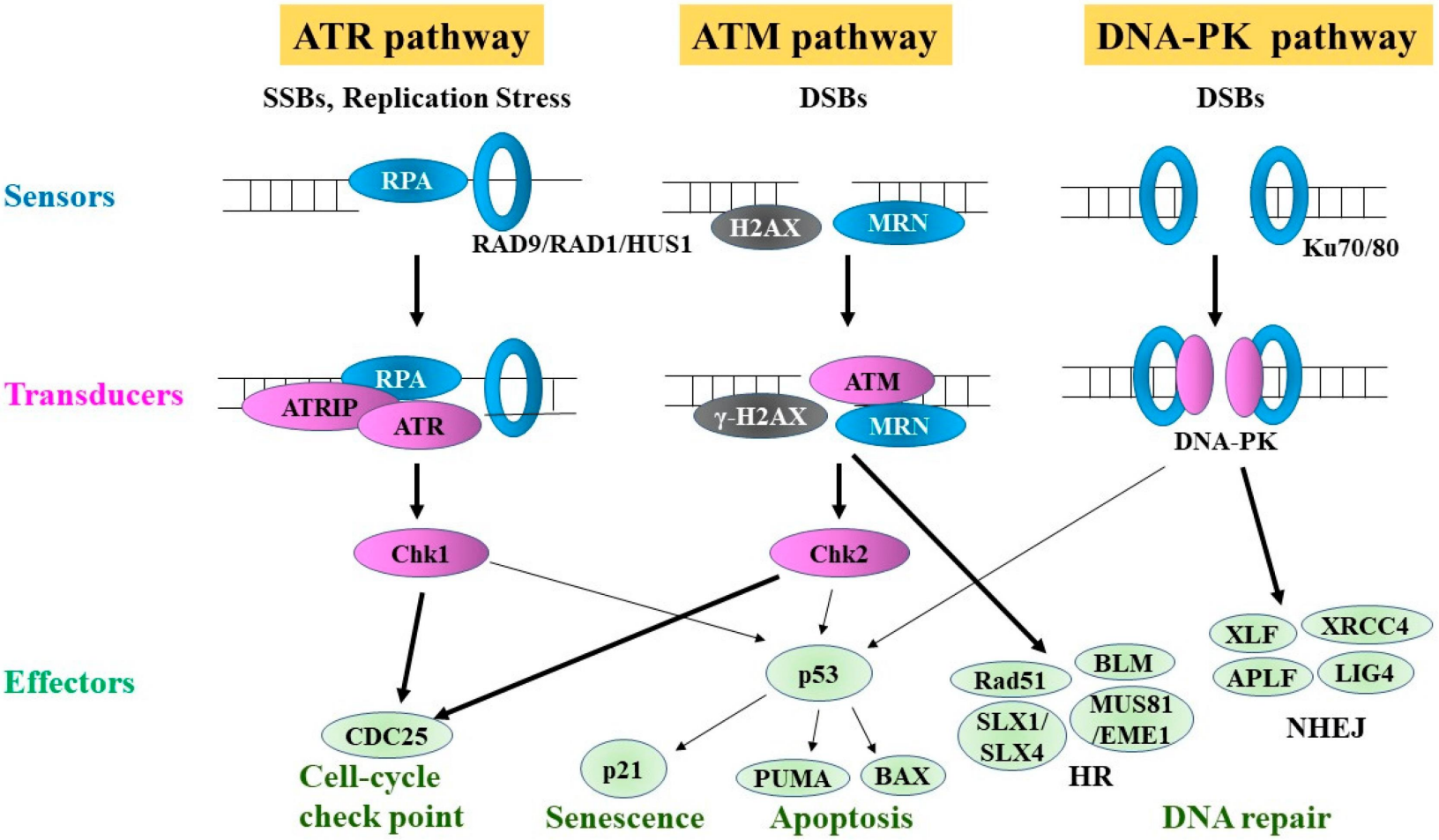
ATM and DNA-PK as the important kinases in the DNA DSB pathway



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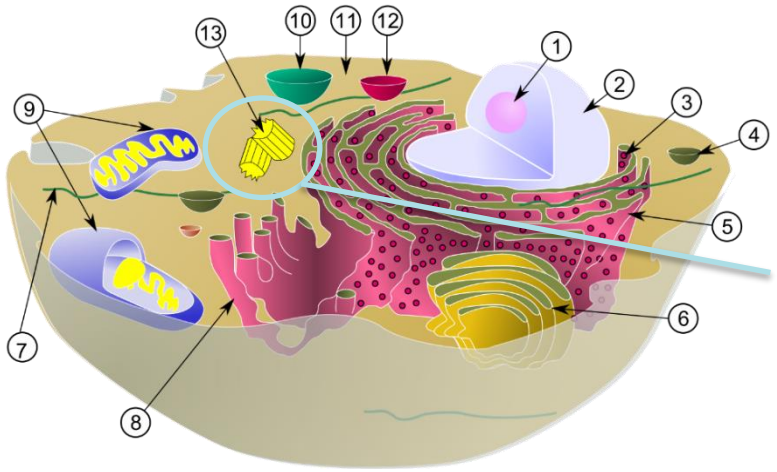
Saitoh et al. *Cancers* 2021

Main cell death pathways in response to ionising radiation



Mitotic catastrophe - cell stress which occurs as a result of aberrant mitosis

- Formation of giant cells with aberrant nuclear morphology
- Centrosome hyperamplification
- Multiple nuclei and/or several micronuclei

Cells may survive for days, transit into senescence, or die by delayed apoptosis or delayed necro(pto)sis

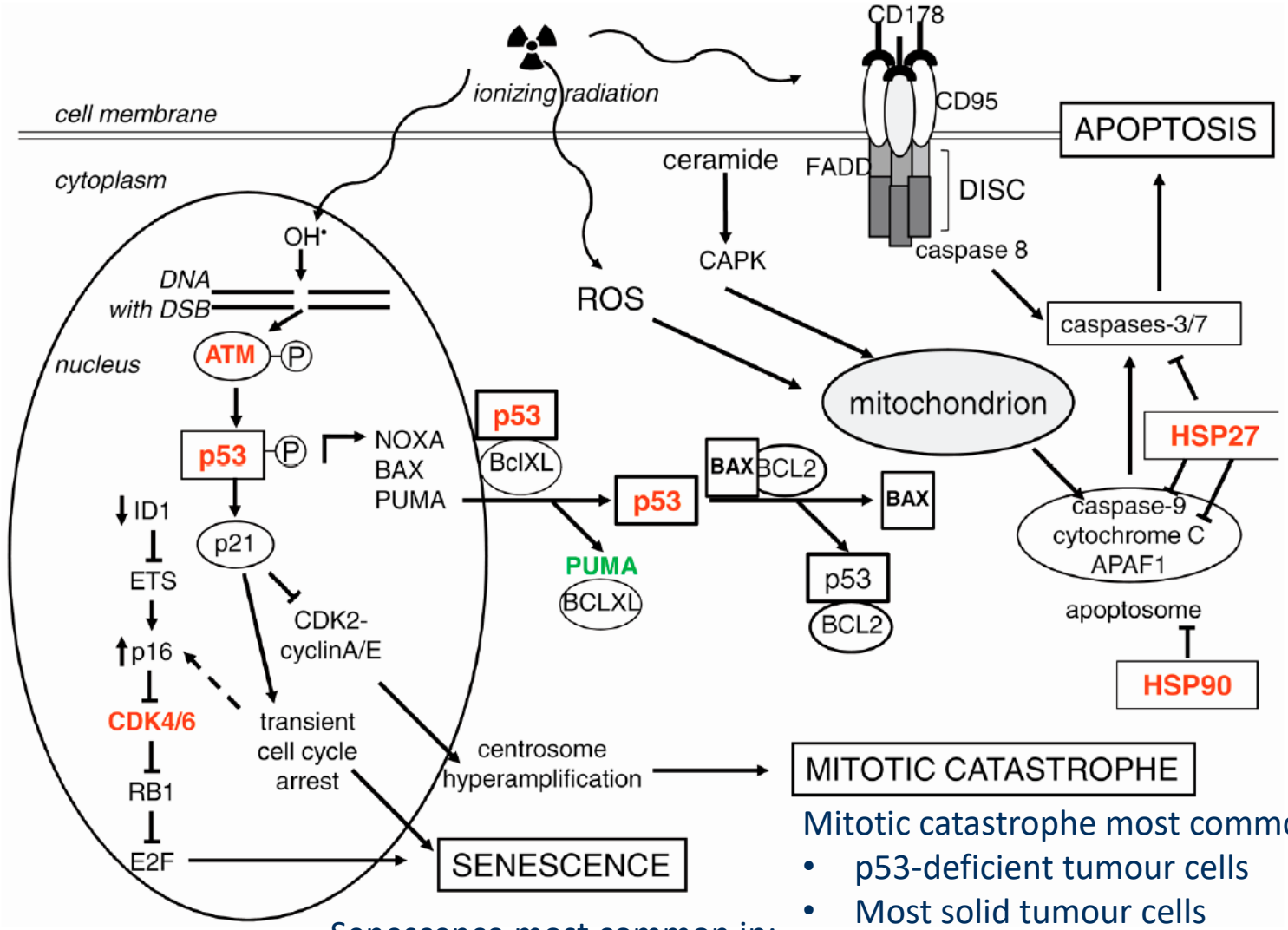
Apoptosis – programmed cell death

- Cellular shrinkage
- Chromatin condensation
- Nuclear fragmentation
- Membrane blebbing
- Markers: PARP cleavage, caspase 3 cleavage, positive staining for annexin V

Senescence - cells exit the cell cycle and do not further undergo cell division, but may remain metabolically active

- Enlarged and flattened cellular morphology, increased granularity
- Upregulation of cyclin-dependent kinase inhibitors, such as p16INK4a, p21Waf1, and p27Kip1
- Marker: Positive staining for the senescence-associated β-galactosidase (SA-β-Gal)

Main cell death pathways – cell type selectivity



Apoptosis most common in:

- Hematopoietic cells
- Gastro-intestinal tract mucosa cells
- p53 wildtype tumour cells

Mitotic catastrophe most common in:

- p53-deficient tumour cells
- Most solid tumour cells

Senescence most common in:

- Normal tissues

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Maier et al, Int J Mol Sci. 2016 Jan; 17(1): 102.

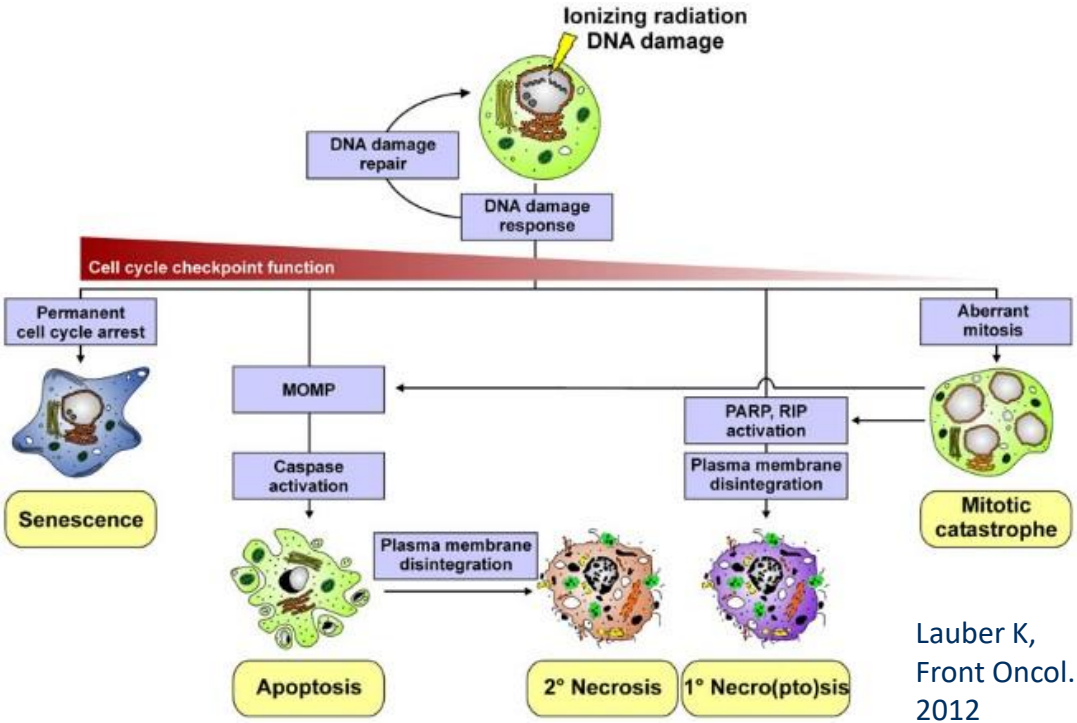
Alternative types of cell death – necrosis, necroptosis

- High single doses during ablative radiotherapy can cause **necrosis**:
  - Accidental, uncontrolled form of cell death as a consequence of excessive physical/chemical stress
  - Causes inflammation

- Radiation-induced DNA damage – especially when combined with hyperthermia - can cause **necroptosis**
  - Hyperactivation of the DNA repair enzyme PARP and depletion of intracellular ATP levels
  - Activation of RIP
  - Production of reactive oxygen species (ROS), lipid peroxidation, swelling of organelles, rupture of the plasma membrane, and release of intracellular contents

MOMP; mitochondrial outer membrane permeabilisation

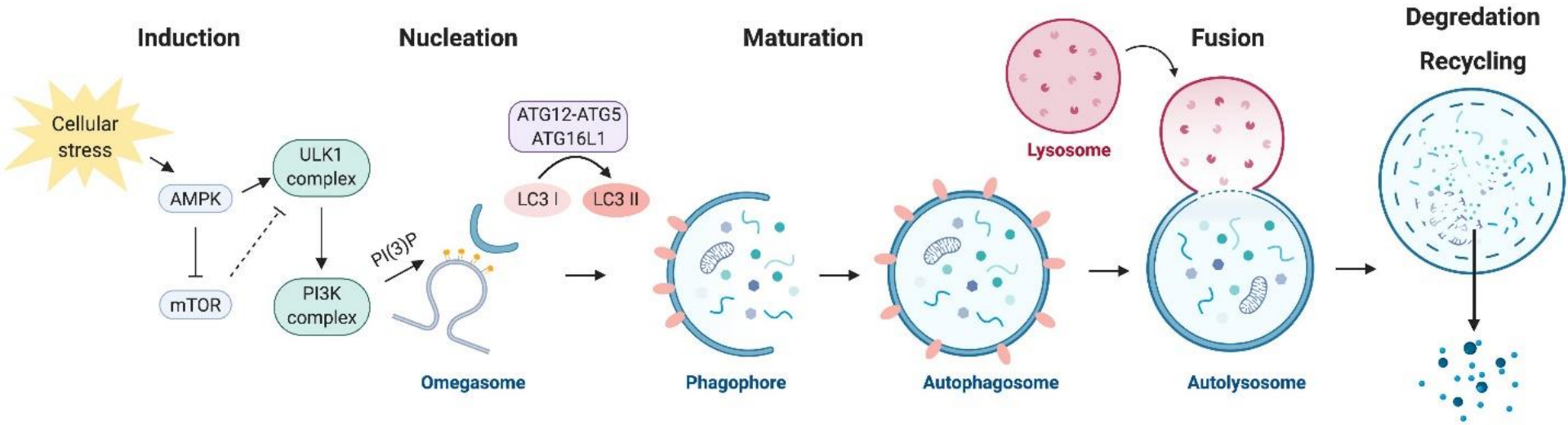
PARP: poly-ADP-ribose-polymerase
RIP: receptor interacting protein



Lauber K, Front Oncol. 2012

Alternative types of cell death – autophagy

- "Cellular self-digestion"
- Normally, cytoplasmic components are encapsulated in autophagosomes, which fuse with lysosomes where material is degraded before recycling
- IR can induce autophagy and cell death via f.ex. ATM or ROS, but may also promote survival



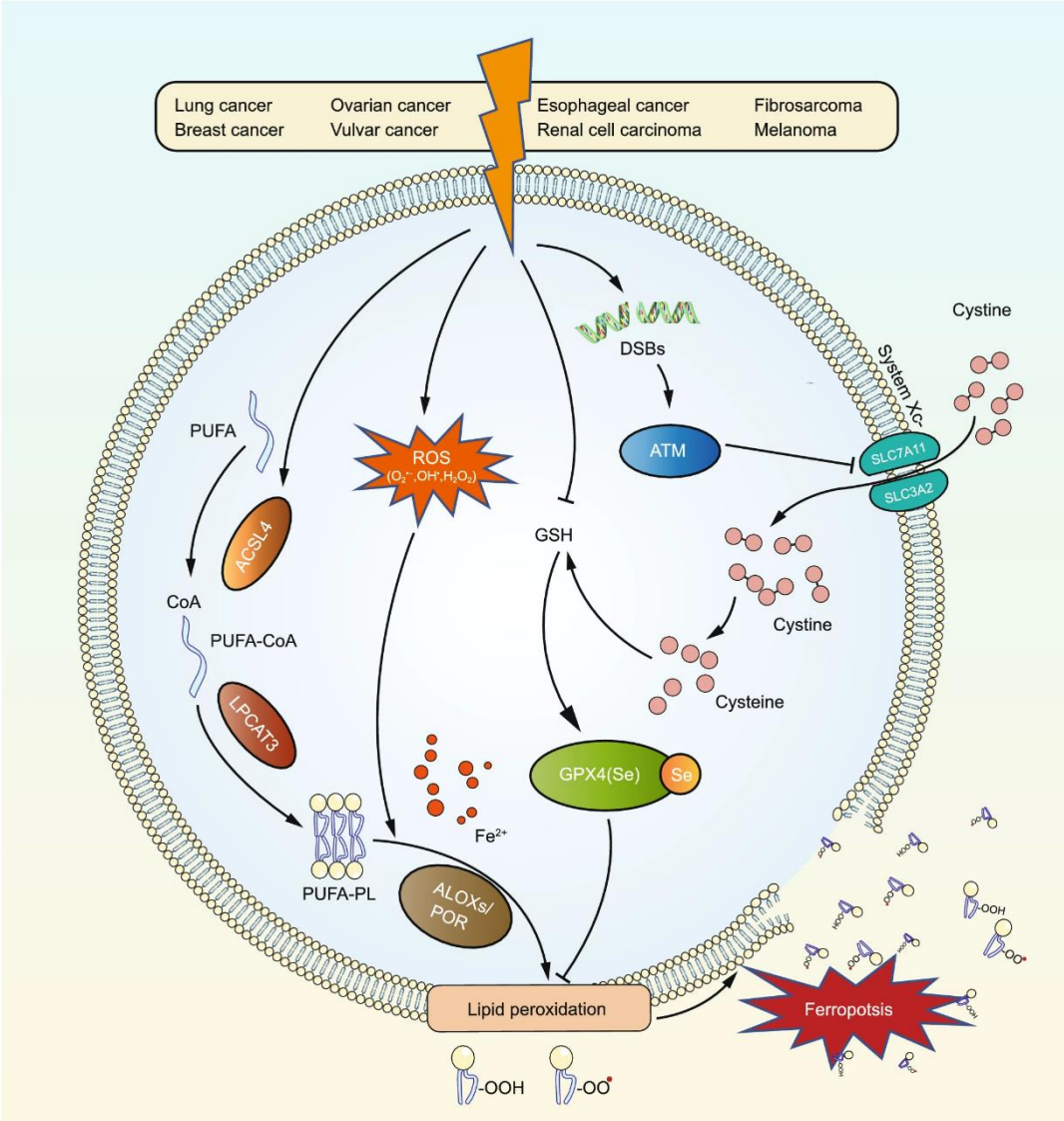
Su et al. Front. Cell Dev. Biol. 2022, Hu et al. Int J Oncol 2016

Chang, Front Cell Dev Biol 2020

Alternative types of cell death – ferroptosis

"Newest cell death mode"

- Driven by iron-dependent phospholipid peroxidation
  - Accumulation of lipid reactive oxygen species (ROS), shrunken mitochondria, membrane integrity damage
- IR can induce ferroptosis by producing ROS - involved in radiation injury in normal cells
- Therapy-resistant cancer cells are more vulnerable to ferroptosis (inducer may act as radiosensitisers)



Lei et al. Prot & Cell 2021

Factors influencing cellular radiosensitivity

- **Physical factors**
  - **Dose, radiation quality, dose rate, fractionation, temperature**
- Chemical factors
  - Oxygen, radiosensitisers, radioprotectors
- Biological factors
  - Organism level: Whole/partial body exposure, age, inherited genetic disorders, inflammatory state/immune response/infections/microbiome
  - Cellular level: Cell cycle stage, stem cell/differentiated cell type, chromatin conformation
- Technical factors
  - Accuracy of radiotherapy delivery

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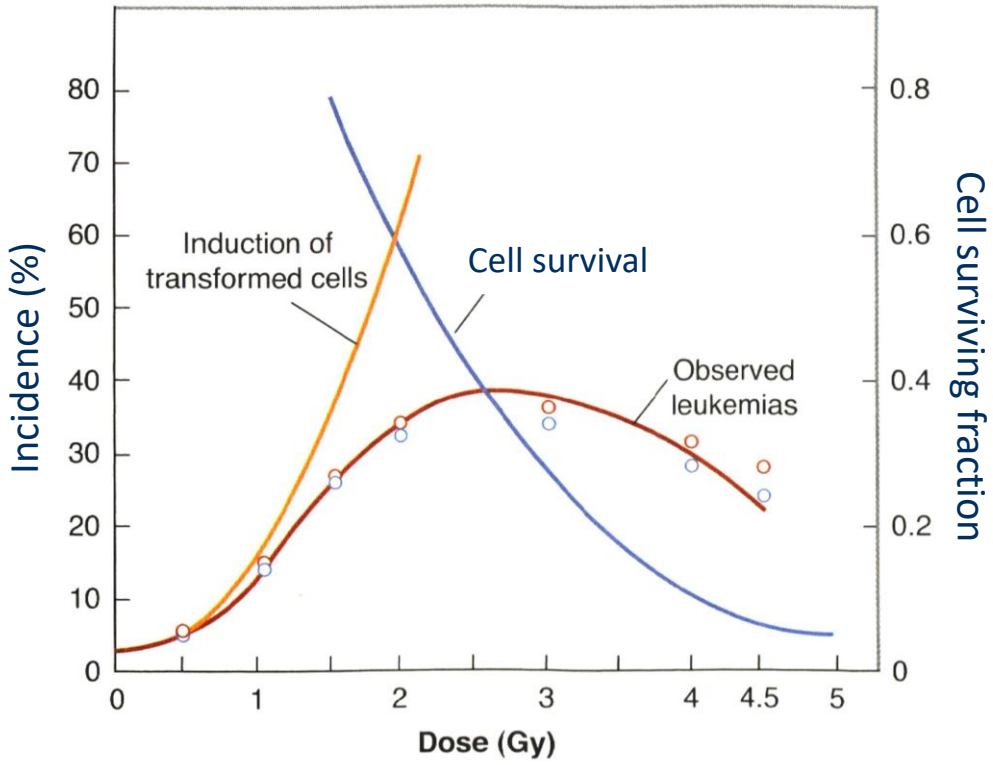
Dose

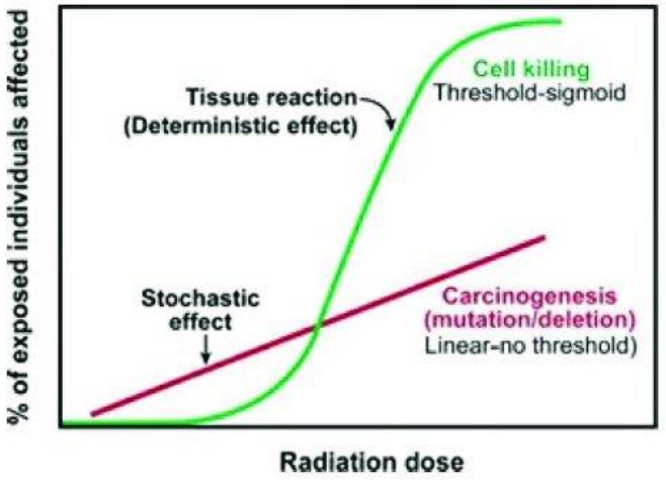
Radiation dose response – Cross-relationship between cell death and mutations



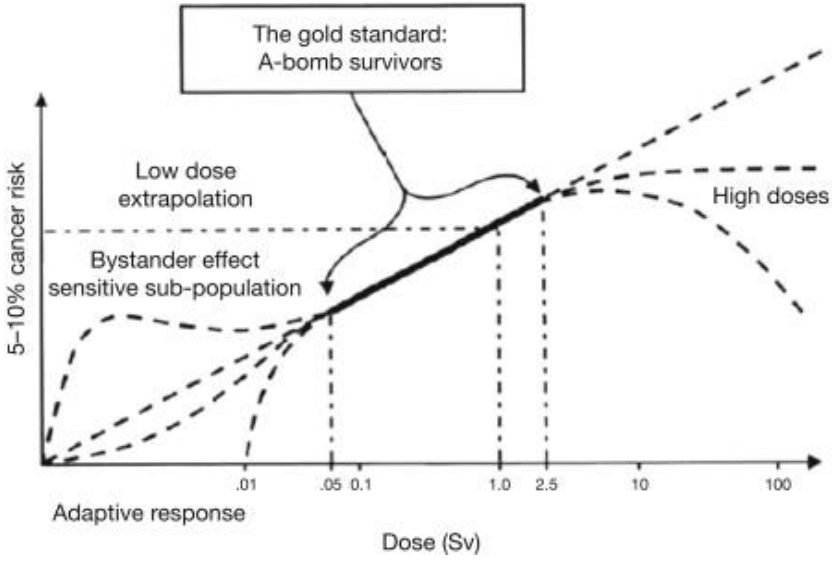
The incidence of radiation-induced leukemia follows a bell shape because of the balance between cell killing and induction of transformed cells

LH Gray, Radiation Biology and Cancer, 1965

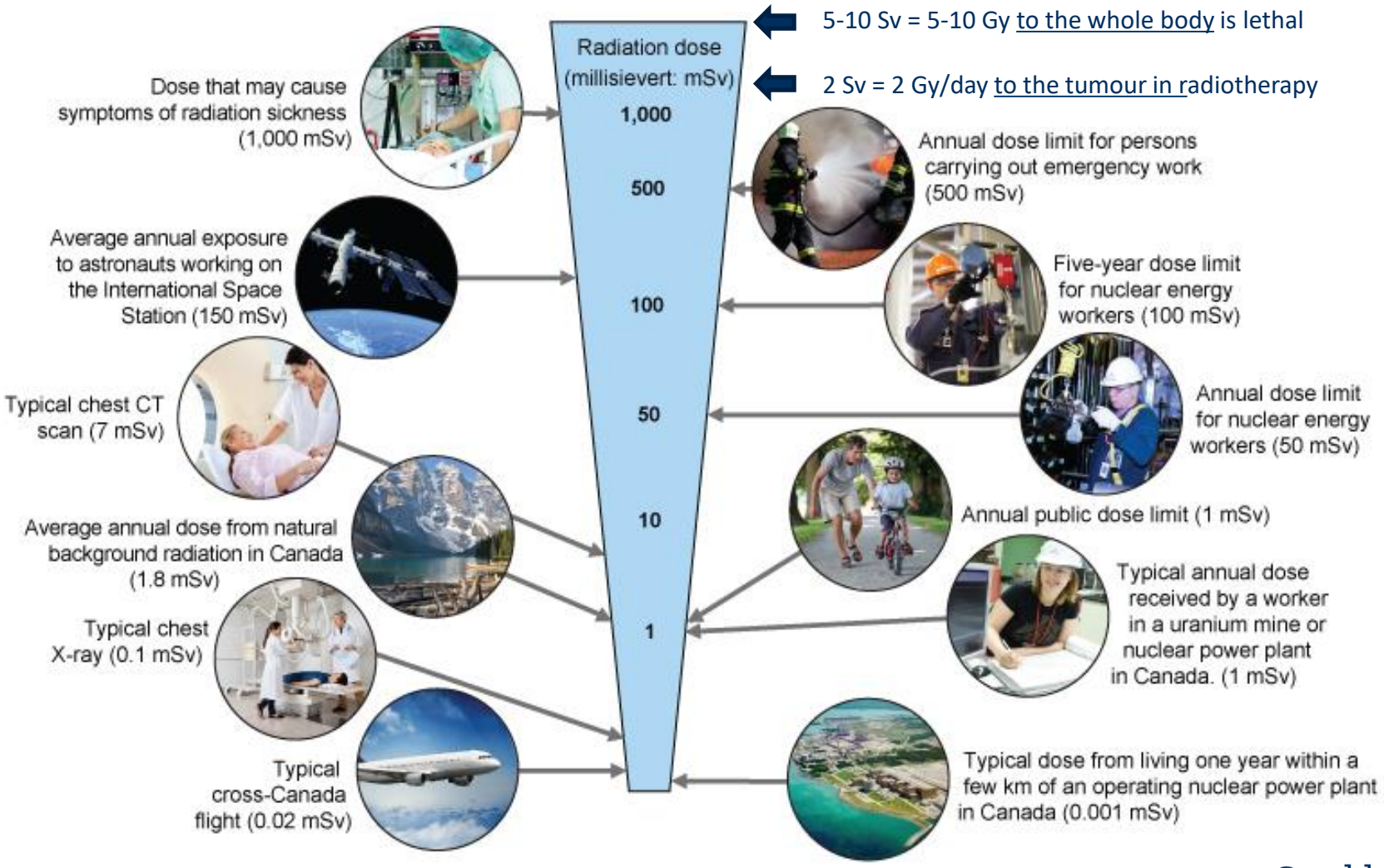
Different patterns for carcinogenesis vs tissue reactions



Uncertainties in the very high and low dose ranges



Radiation exposure occurs at a range of doses



Worldwide average effective dose from natural radiation: 2.4 mSv/year

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nuclearsafety.gc.ca

Dose ranges

- High: >1 Gy
  - radiotherapy or radiological accidents
- Moderate: 100 mGy to 1 Gy
  - e.g. Chernobyl accident recovery operation workers
- Low: 10 to 100 mGy
  - Multiple computerized tomography (CT) scans
- Very low: <10 mGy
  - Single CT or conventional radiology without CT or fluoroscopy

- For high LET radiation:
  - ≤ one track traversal per cell is considered low, but note that a low dose is not a reality per cell, instead it means a low likelihood of cells being hit

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UNSCEAR 2012 Report

Medical exposure

	Procedure	Approximate effective radiation dose	Comparable to natural background radiation for:
NUCLEAR MEDICINE	Positron Emission Tomography–Computed Tomography (PET/CT)	25 mSv	8 years
HEART	Coronary Computed Tomography Angiography (CTA)	12 mSv	4 years
ABDOMINAL REGION	Computed Tomography (CT)–Abdomen and Pelvis	10 mSv	3 years
CHEST	Computed Tomography (CT)–Chest	7 mSv	2 years
CENTRAL NERVOUS SYSTEM	Computed Tomography (CT)–Head	2 mSv	8 months
CHEST	Spine X-ray	1.5 mSv	6 months
BREAST	Mammography	0.4 mSv	7 weeks
BONE	Chest X-ray	0.1 mSv	10 days
DENTAL	Dental X-ray	0.005 mSv	1 day
BONE	Extremity (hand, foot, etc.) X-ray	0.001 mSv	3 hours
BONE	Bone Densitometry (DEXA)	0.001 mSv	3 hours

Doses acquired from computed tomography (CT) or X-rays (equal to time of natural background levels)

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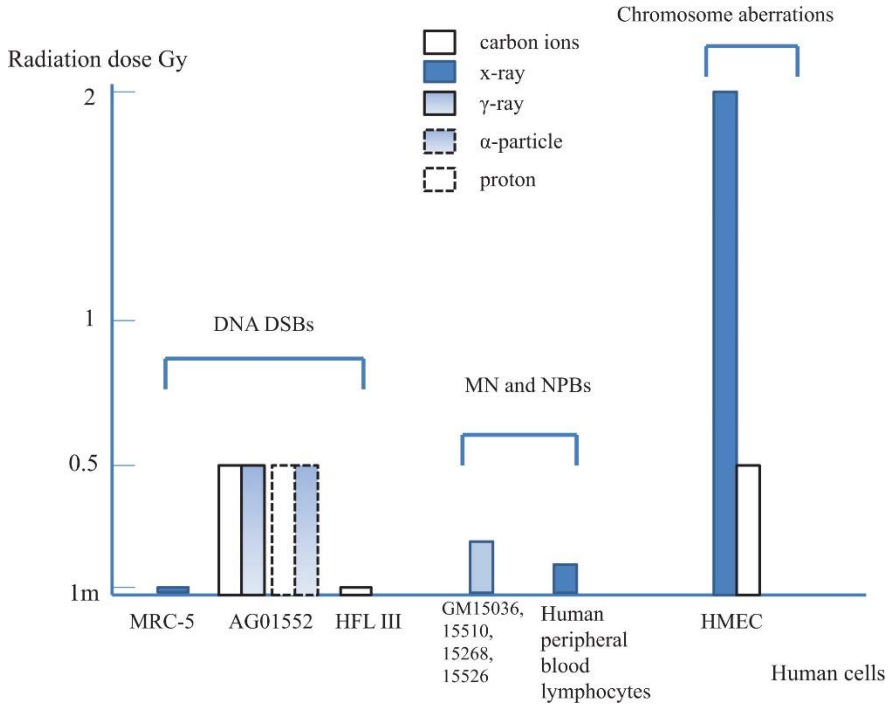
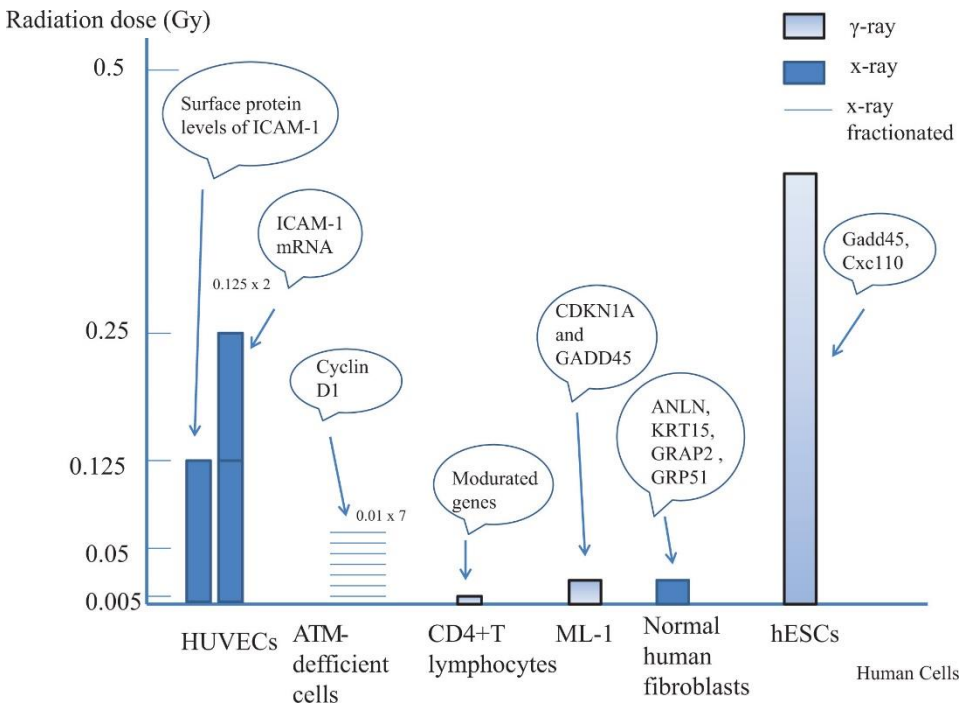
www.radiologyinfo.org

Which are the lowest radiation doses giving measurable effects?

- A linear correlation exist between radiation dose and
  - number of γH2AX foci: down to 1 mGy
  - chromosomal aberrations: >20 mGy
- Generally, gene expression/stress responses can be detected after lower doses than DNA DSB or chromosomal damage, but responses are always cell model-, radiation quality- and scheme-dependent





Shimura et al. Dose response 2018

Inefficient DNA damage response exerted by low doses of radiation?

- After 20-80 mGy, the γH2AX foci did not decrease, phospho-ATM did not colocalise with γH2AX foci, proliferation remained
  - Inefficient repair or new γH2AX appear from replication stress

- A threshold dose (0.2-0.6 Gy/10-20 DSBs, depending on cell type) has been suggested below which ATM-dependent, early G2/M arrest is not activated
  - Possible for cells with unrepaired DSBs to enter mitosis, which might result in loss of genetic material

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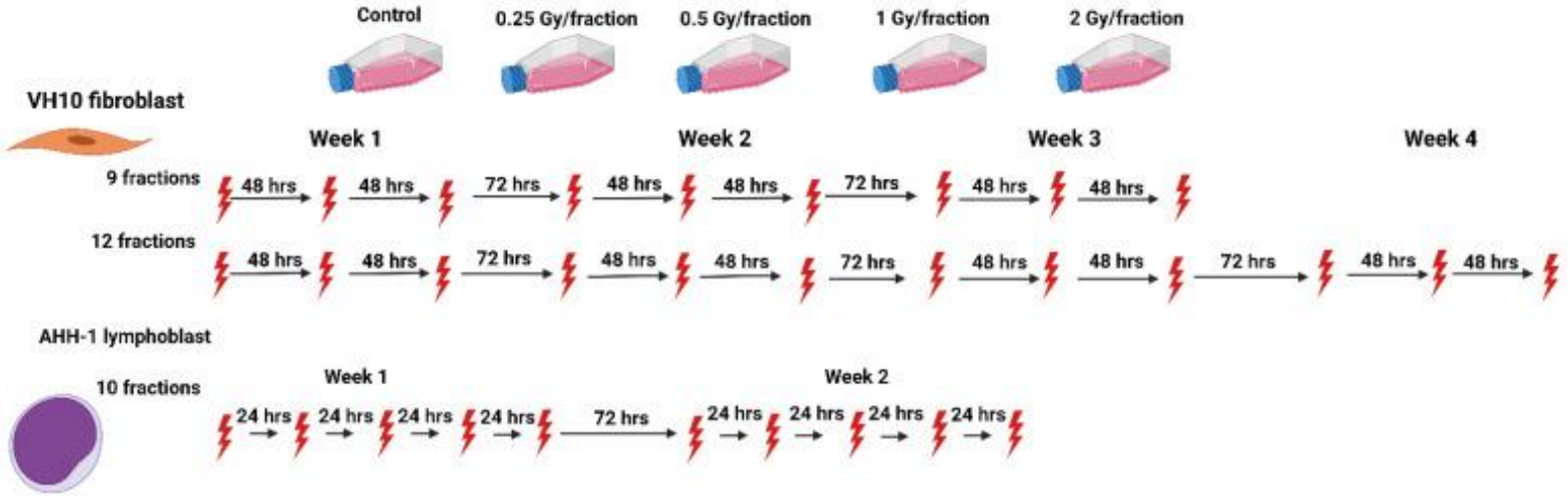
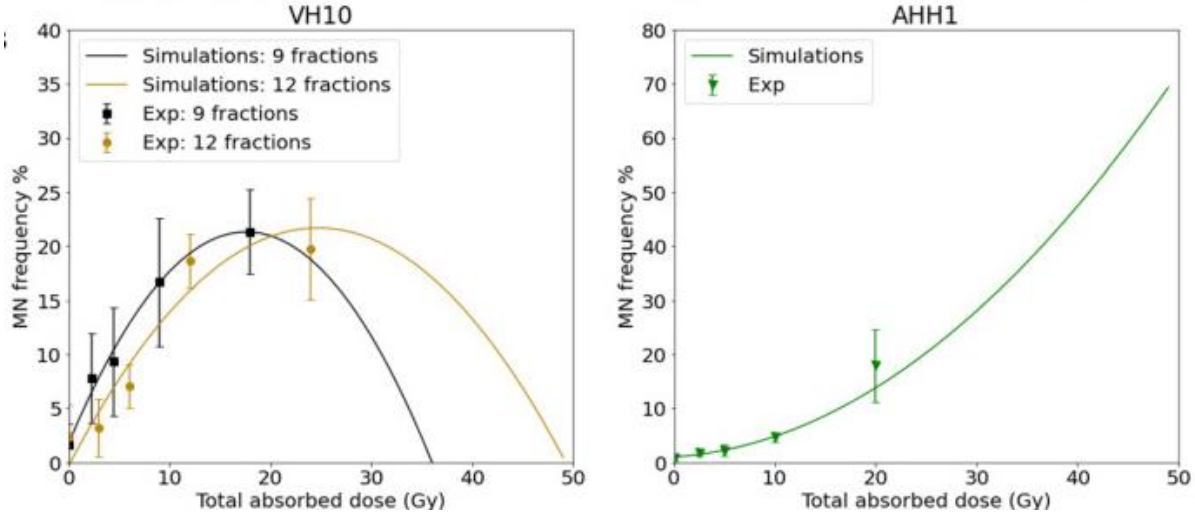
Piotrowski et al. Radiol Oncol 2017

What happens at high fractionated doses in normal cells?



Cell-type dependent patterns for MN at 3 days post fractionated gamma radiation, which may be due to their cell death modes (fibroblasts prone to senescence, lymphoblasts to apoptosis)



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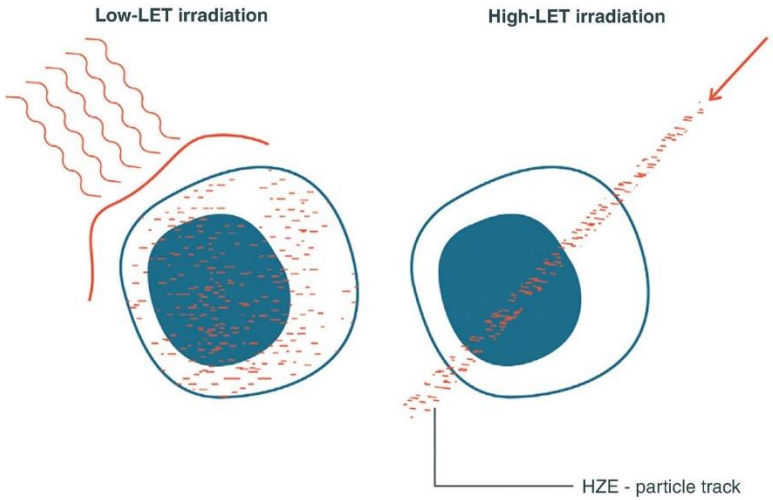
Akuwudike et al. Int J Mol Sci. 2022

HZE = high Z element/high atomic number

Radiation quality

Sparsely ionising
low LET radiation

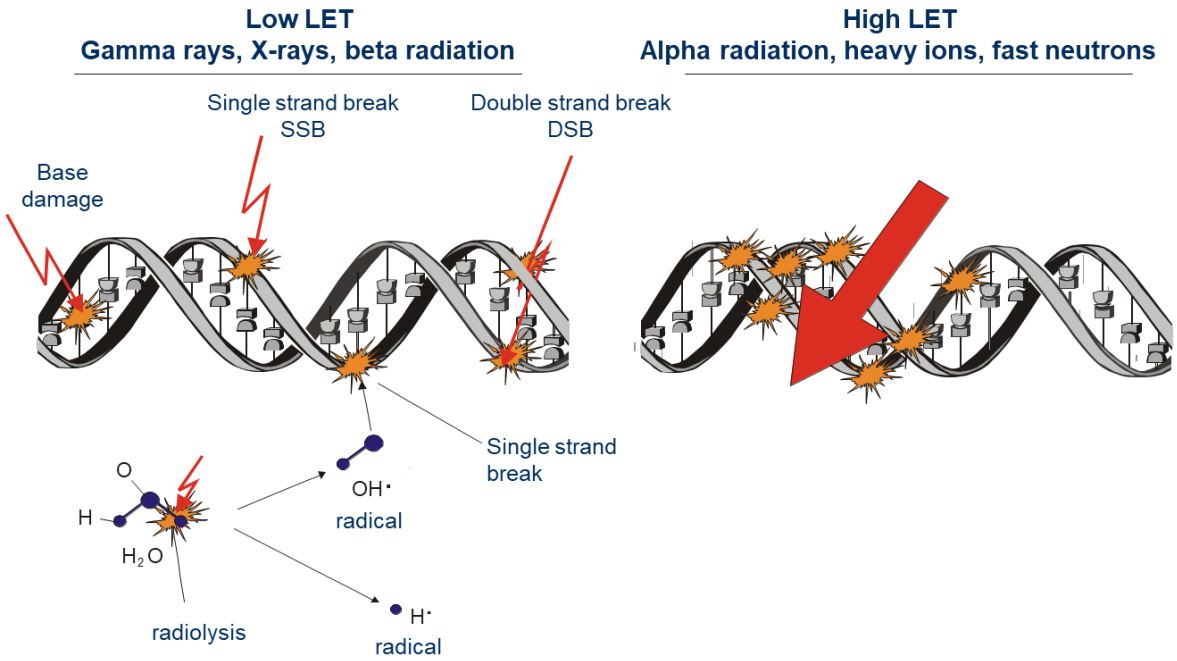
<10 keV/µm



Densely ionising
high LET radiation

>10 keV/µm

LET: Linear Energy Transfer
The energy transferred per unit path length traversed by an ionising particle



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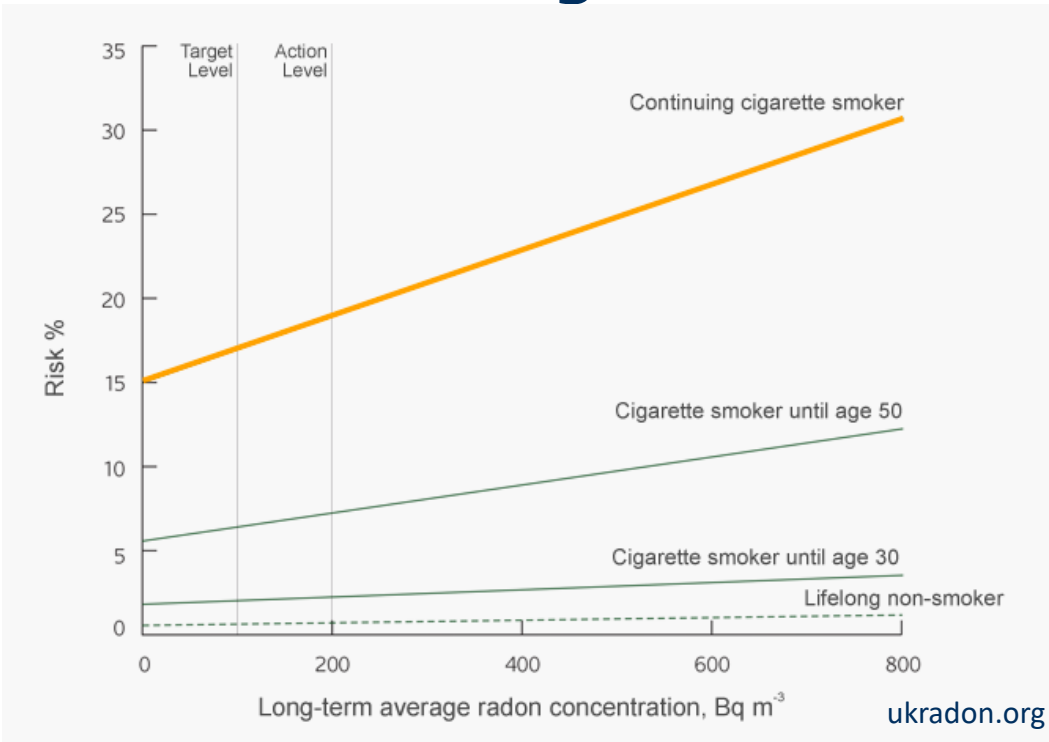
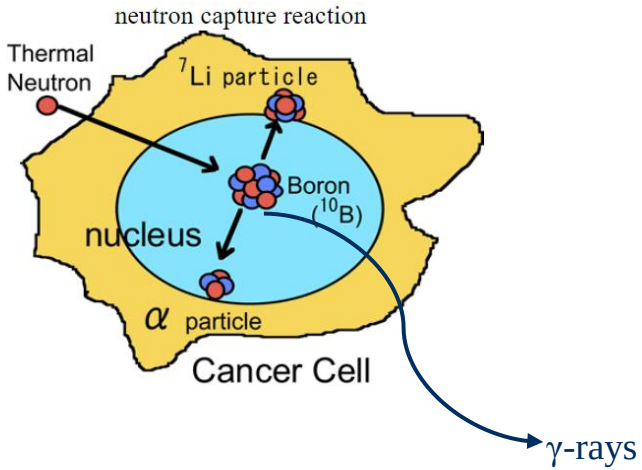
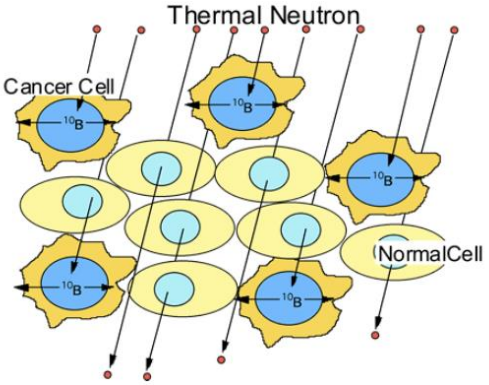
Moreels et al. 2019

Relevance of a higher LET for biological responses

- **Radioprotection**
  - Radon (^{86}Rn, alpha radiation)
  - Cosmic radiation (protons, alpha radiation)
- **Radionuclide therapy**
  - Alpha emitters, ^{223}Ra for castrate-resistant prostate cancer
- **Radiotherapy**
  - Carbon ions
  - Boron neutron capture therapy (BNCT)







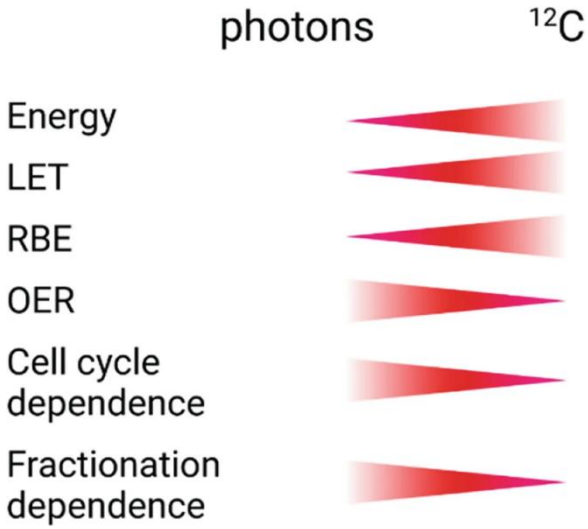
Kawabata et al., 2008

Response to high LET compared to low LET DNA damage

Clustered DNA damage - Two or more lesions formed within one or two helical turns of DNA caused by the passage of a single radiation track (Ward 1994)

High LET

- Causes more complex damage
  - DSB-related - DSB are surrounded by other lesions
  - Non-DSB oxidative clustered DNA lesions - DSB are not involved
- Slower kinetics of DNA repair
- Less dependent chromatin structure, oxygen levels and cell cycle



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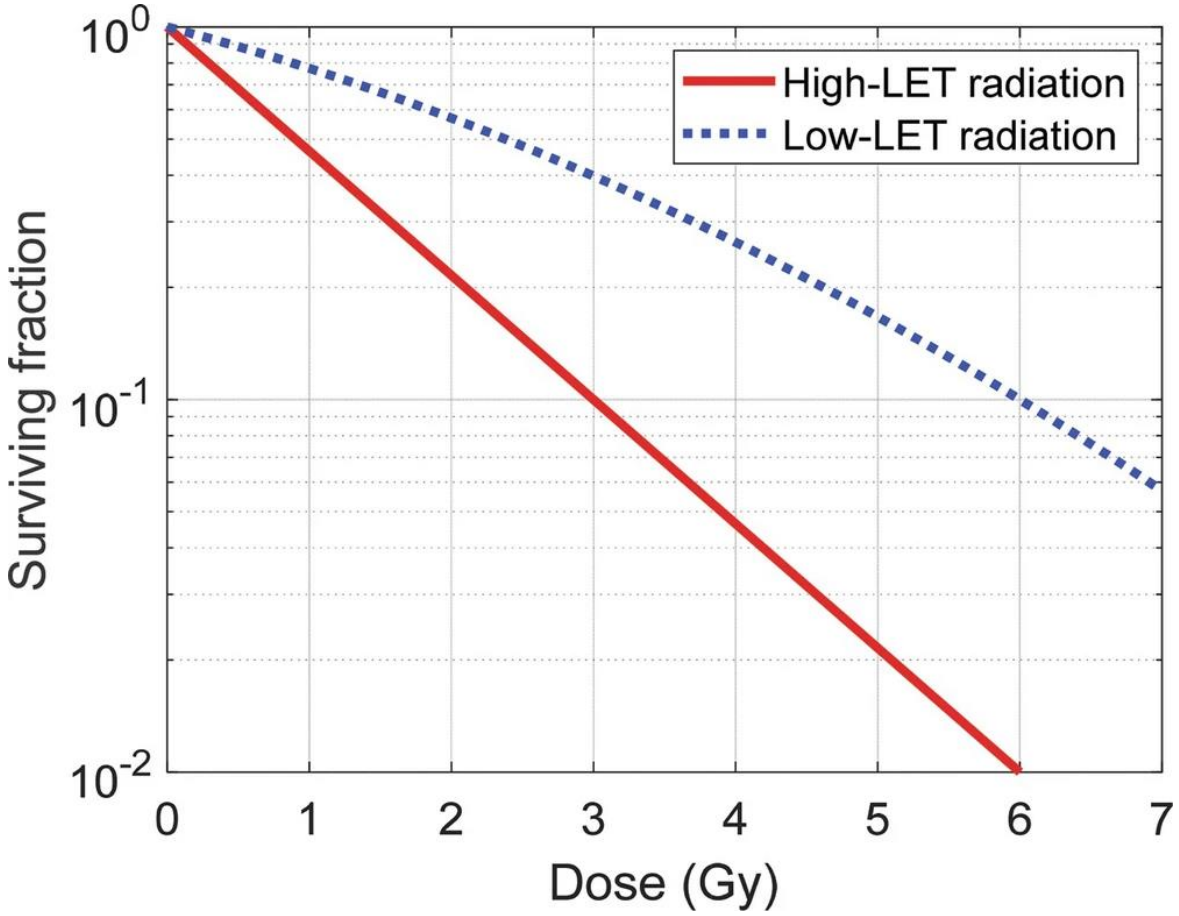
Baatout et al. ed. 2023. Radiobiology Textbook

Higher relative biological effectiveness (RBE) of high vs low LET



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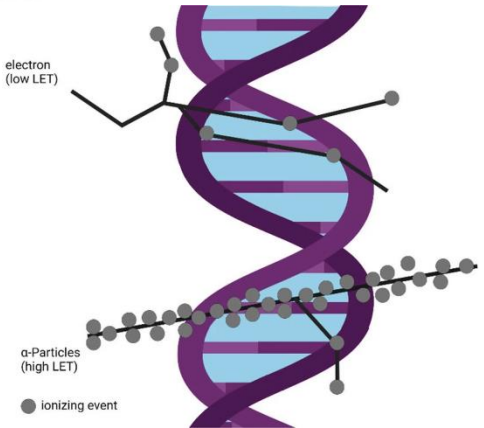
Greater mutagenic and cytotoxic effects of clustered lesions compared to isolated lesions

Mutagencity / Cytotoxicity

Isolated single-strand damage	Clustered single-strand damage	Clustered double-strand damage	Clustered DSB + single-strand damage	Clustered DSBs (+/- other lesions)
Base lesion	Base damage + SSB	Base damage + SSB	Base damage + DSB	Two DSBs
Missing base	Multiple base lesions	Multiple base lesions	SSB + DSB	DSBs + SSB
SSB				

Reparabilty

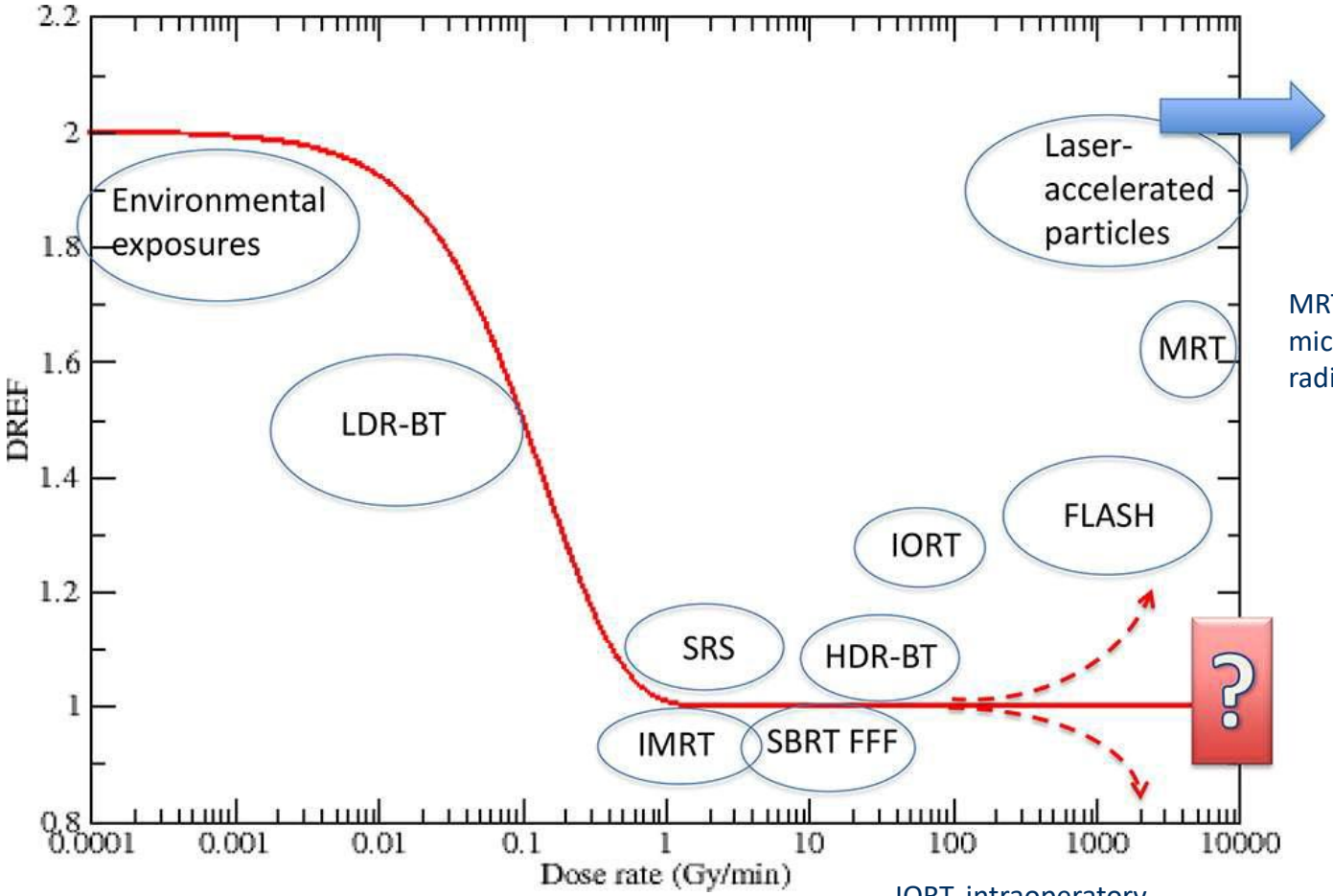
Higher LET →



Nickoloff et al. 2020
Baatout et al. ed. 2023. Radiobiology Textbook

Dose rate (dose delivery per unit time)

Exposure occurs at a range of dose rates



MRT, microbeam radiotherapy

IORT, intraoperatory radiotherapy

IMRT, intensity modulated radiotherapy

SRS, stereotactic radiosurgery

LDR-BT, low dose-rate brachytherapy

HDR-BT, high dose-rate brachytherapy

SBRT-FFF, stereotactic body radiotherapy flattening filter free

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Durante et al. Br J Radiol 2018

How to take dose rate into account?

- A low dose rate (LDR) is defined as
  - ≤ 0.1 mGy/min for low LET radiation
  - one-track traversal per cell per hour for high LET radiation

- Dose-rate effectiveness factor (DREF)
  - Is chronic radiation exposure protective compared to acute exposure?
  - Different radiation protection organisations recommend different factors to divide by to estimate the cancer risk - DREF: 2 / 1.5 / 1
  - A DREF (DDREF) of 2 → risk reduced by half by chronic exposure

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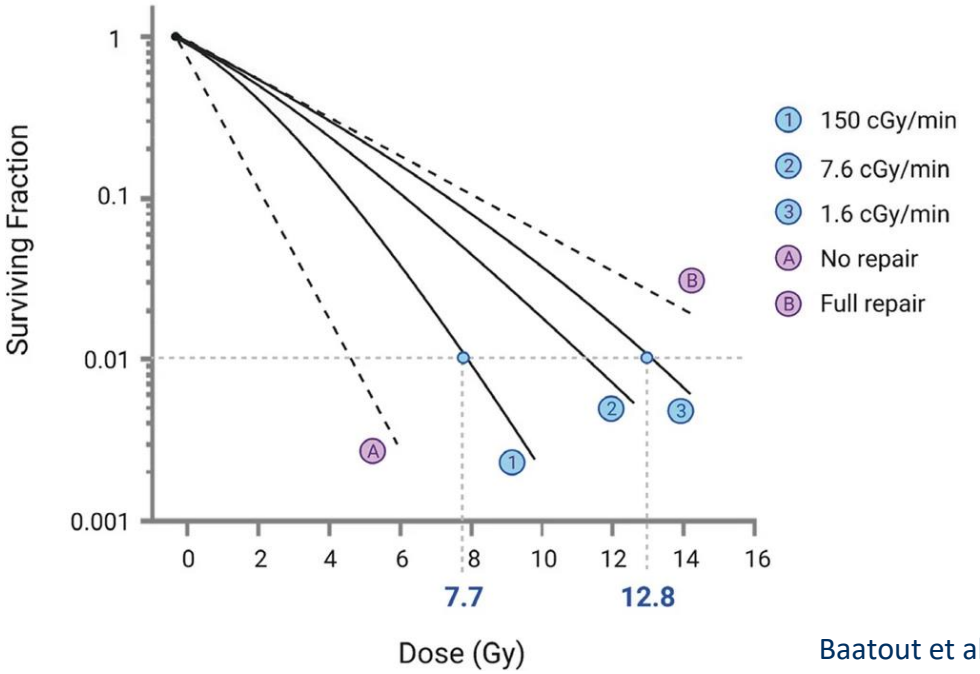
Dose rate effect

- In many cases, a dose rate effect can be seen
  - Decrease in biological effectiveness by a lower dose rate
  - Commonly attributed to repair occurring during exposure
- Sometimes, an inverse dose rate effect can be seen
  - Increase in biological effectiveness by a lower dose rate
  - Can be attributed to progression of cells to the more radiosensitive G2 cell cycle phase



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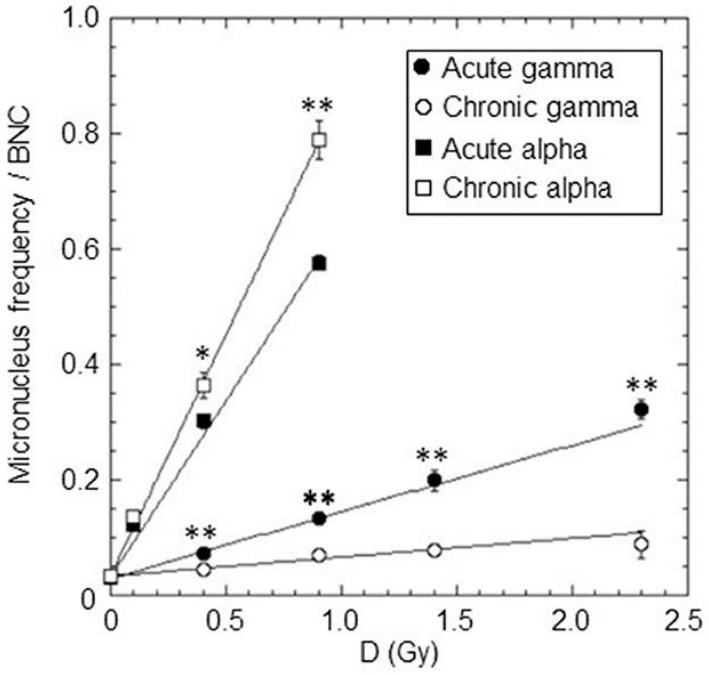
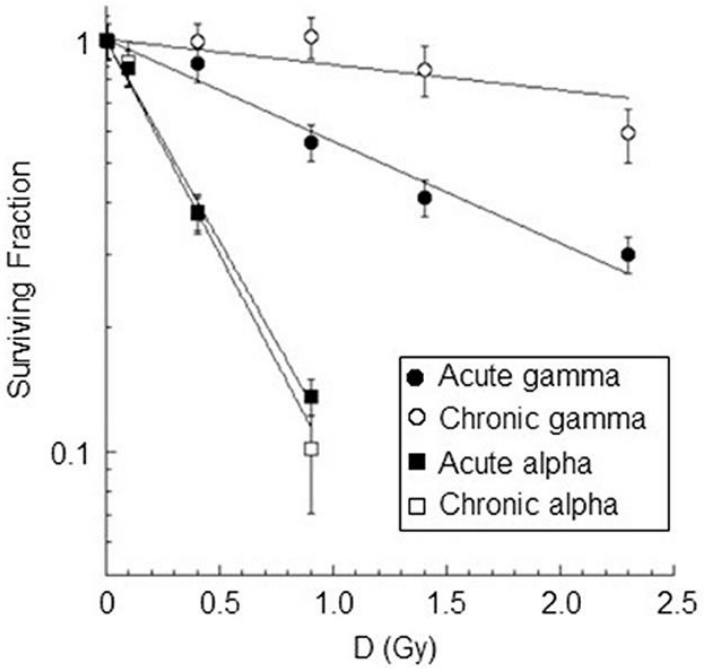
Opposing effects of low dose rate on cytogenic damage for gamma and alpha radiation





Confluent human fibroblasts (AG1522 cells) irradiated at a high dose rate (40,200 and 4,980 mGy/h for gamma rays and alpha particles, respectively) and at a low dose rate (~18 mGy/h for both gamma rays and alpha particles)

X-rays: Reduced cytogenetic damage and higher clonogenic cell survival when the dose was delivered chronically instead of acutely

Alpha particles: Greater cytogenetic damage for chronic exposure (≥ 0.4 Gy) and equal reduction of clonogenic cell survival for both chronic and acute exposure

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Anello et al. Front Publ Health 2024

Ultra-high dose-rate

- FLASH radiotherapy >40 Gy/s vs 0.02 Gy/s (1 Gy/min)
- Relative protection of normal tissues compared with conventional dose rate radiotherapy is suggested
  - All local oxygen used up in fully oxic normal cells – creating transient radioresistance
  - Generally hypoxic tumour cells - similar effects as conventional RT
  - Eliminate motion effects, provided targeting is well controlled
- Promising data from various animal models
- First *in human* FLASH-RT treatment was feasible and safe and favorable both on normal skin and the tumour
- **Hypoxia effect only or also ”pure” dose rate effect?**

Long-term neurocognitive benefits of FLASH radiotherapy driven by reduced reactive oxygen species

Pierre Montay-Gruel[a,b,1], Munjal M. Acharya[c,1], Kristoffer Petersson[a,b,d], Leila Alikhani[c], Chakradhar Yakkala[a,b], Barrett D. Allen[c], Jonathan Ollivier[a,b], Benoit Petit[a,b], Patrik Gonçalves Jorge[a,b,d], Amber R. Syage[c], Thuan A. Nguyen[c], Al Anoud D. Baddour[c], Celine Lu[c], Paramvir Singh[c], Raphael Moeckli[d], François Bochud[d], Jean-François Germond[d], Pascal Froidevaux[d], Claude Bailat[d], Jean Bourhis[a,b], Marie-Catherine Vozenin[a,b,2,3], and Charles L. Limoli[c,2,3]

Ultra high dose rate (35 Gy/sec) radiation does not spare the normal tissue in cardiac and splenic models of lymphopenia and gastrointestinal syndrome

Bhanu Prasad Venkatesulu[1,6], Amrish Sharma[1,6], Julianne M. Pollard-Larkin[3], Ramaswamy Sadagopan[3], Jessica Symons[1,4], Shinya Neri[1], Pankaj K. Singh[1], Ramesh Tailor[3], Steven H. Lin[1,2,4*] & Sunil Krishnan[1,2,4,5*]

Bourhis et al. 2019, Montay-Gruel et al 2019, Durante et al. 2018, Venkatesulu et al. 2019

Fractionation

Fractionated radiotherapy is based on the 4 Rs

- Repair
  - Tumour cells proliferate faster - have less time to repair the DNA damage before they enter mitosis → mitotic catastrophy
- Redistribution
  - Cell cycle arrest in the relatively radiosensitive G2 phase - next fraction hits those, leading to a high level of cell death
- Reoxygenation
  - First, radiation kills normoxic, proliferating tumour cells. As these die, hypoxic cells move towards blood vessels and become normoxic (therefore weekend breaks) and radiosensitive
- Repopulation
  - The fractionation scheme should ideally be adjusted to the proliferation and kinetics of tumour cells so that repopulation is prevented.
    In some healthy tissues repopulation is so fast that tissue damage is prevented.

A typical clinical radiotherapy regimen:

2 Gy/day, 5 days/week for 6 weeks = 60 Gy

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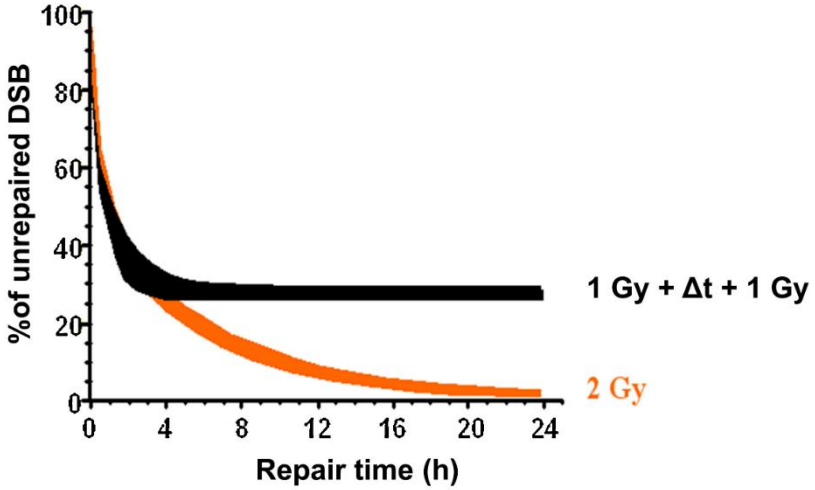
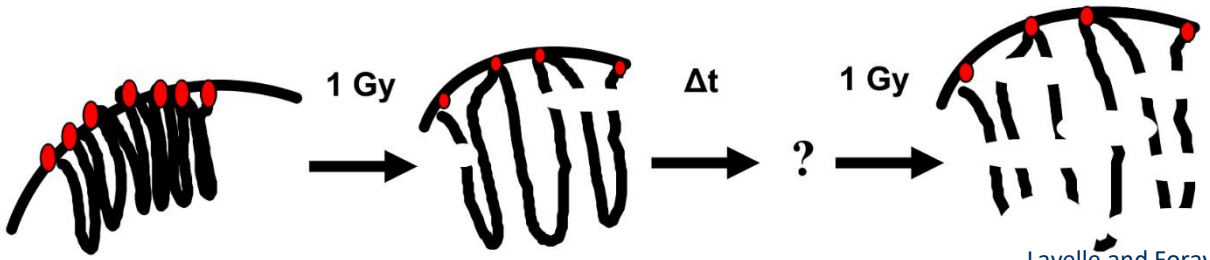
Hall EJ, Giaccia AJ 2012

Fractionated/repeated doses are more problematic for the cell to repair

- Δt is important in the context of chromatin structure, since it takes 12-24 h for the chromatin to rejoin Belyaev et al. Rad Res. 2001
- A more open chromatin is more susceptible to gamma radiation





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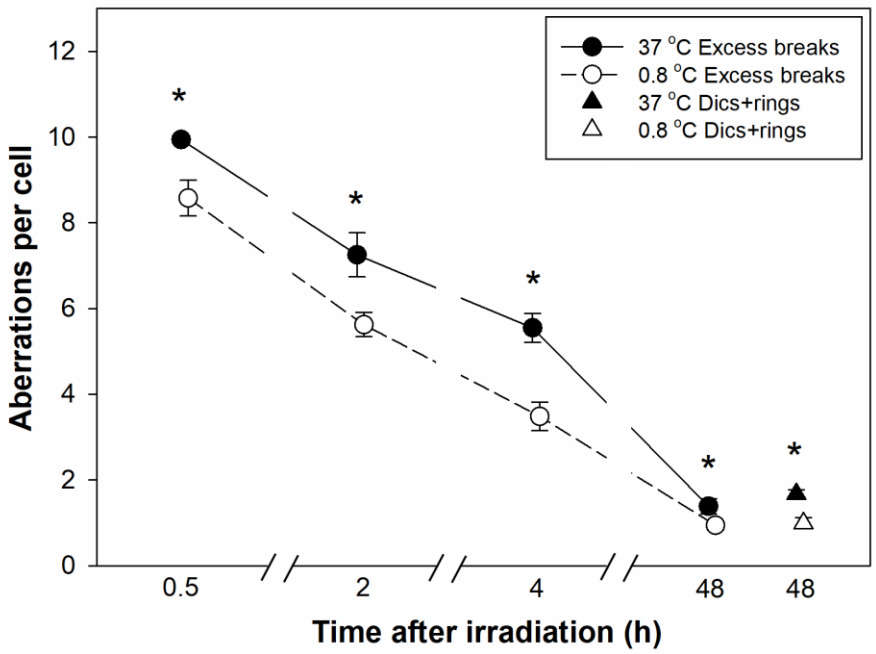
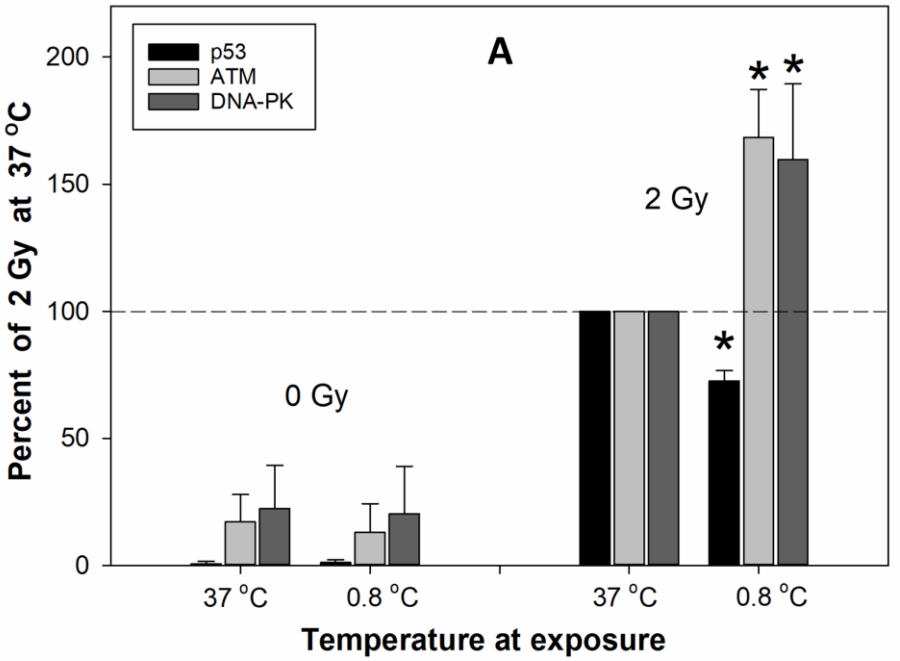
Lavelle and Foray. Int J Biochem Cell Biol. 2014

Temperature (in vitro)

Lower level of chromosomal aberrations and higher activation of DNA damage response proteins when cells are irradiated in ice water (0.8°C)





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Lisowska H et al. Int J Rad Biol 2018

Factors influencing cellular radiosensitivity

- Physical factors
  - Dose, radiation quality, dose rate, fractionation, temperature
- **Chemical factors**
  - **Oxygen, radiosensitisers, radioprotectors**
- Biological factors
  - Organism level: Whole/partial body exposure, age, inherited genetic disorders, inflammatory state/immune response/infections/microbiome
  - Cellular level: Cell cycle stage, stem cell/differentiated cell type, chromatin conformation
- Technical factors
  - Accuracy of radiotherapy delivery

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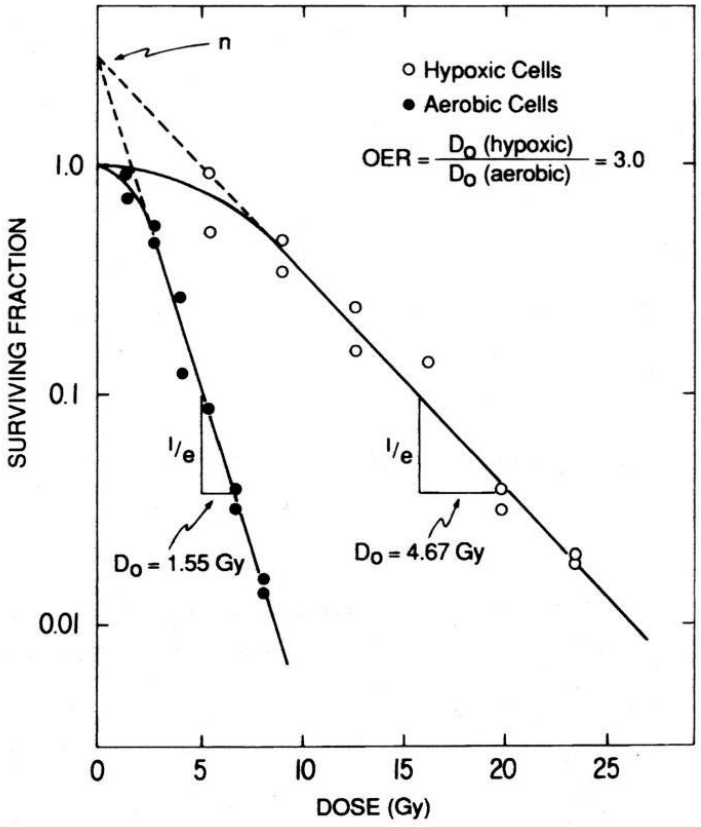
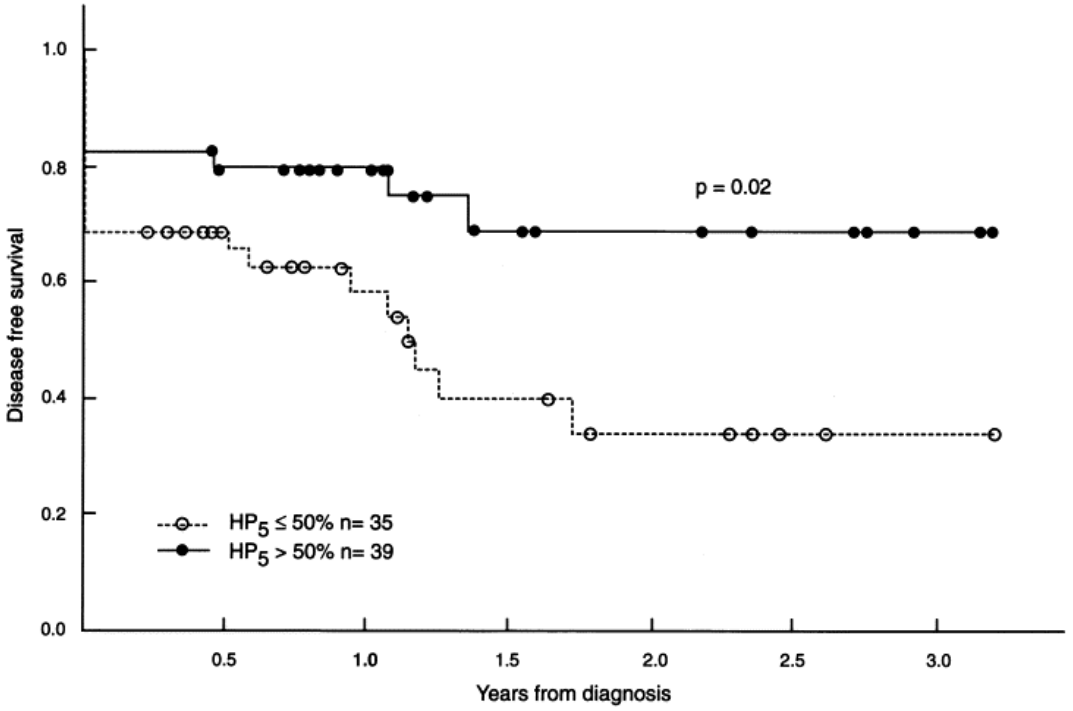
Oxygen





Oxygen potentiates the indirect effect of radiation (via reactive oxygen species)

EMT6 mouse mammary tumour cells were irradiated under aerobic conditions or were made severely hypoxic just before and during irradiation with 250 kV x-rays

Rockwell S, Curr Mol Med. 2009 May; 9(4): 442–458.

Oxygenation predicts radiation response and survival in patients with cervix cancer

Hypoxic proportion HP_5: Percentage of pO_2 readings of <5 mm Hg

Fyles A, Radiotherapy and Oncology 48 (1998) 149–156

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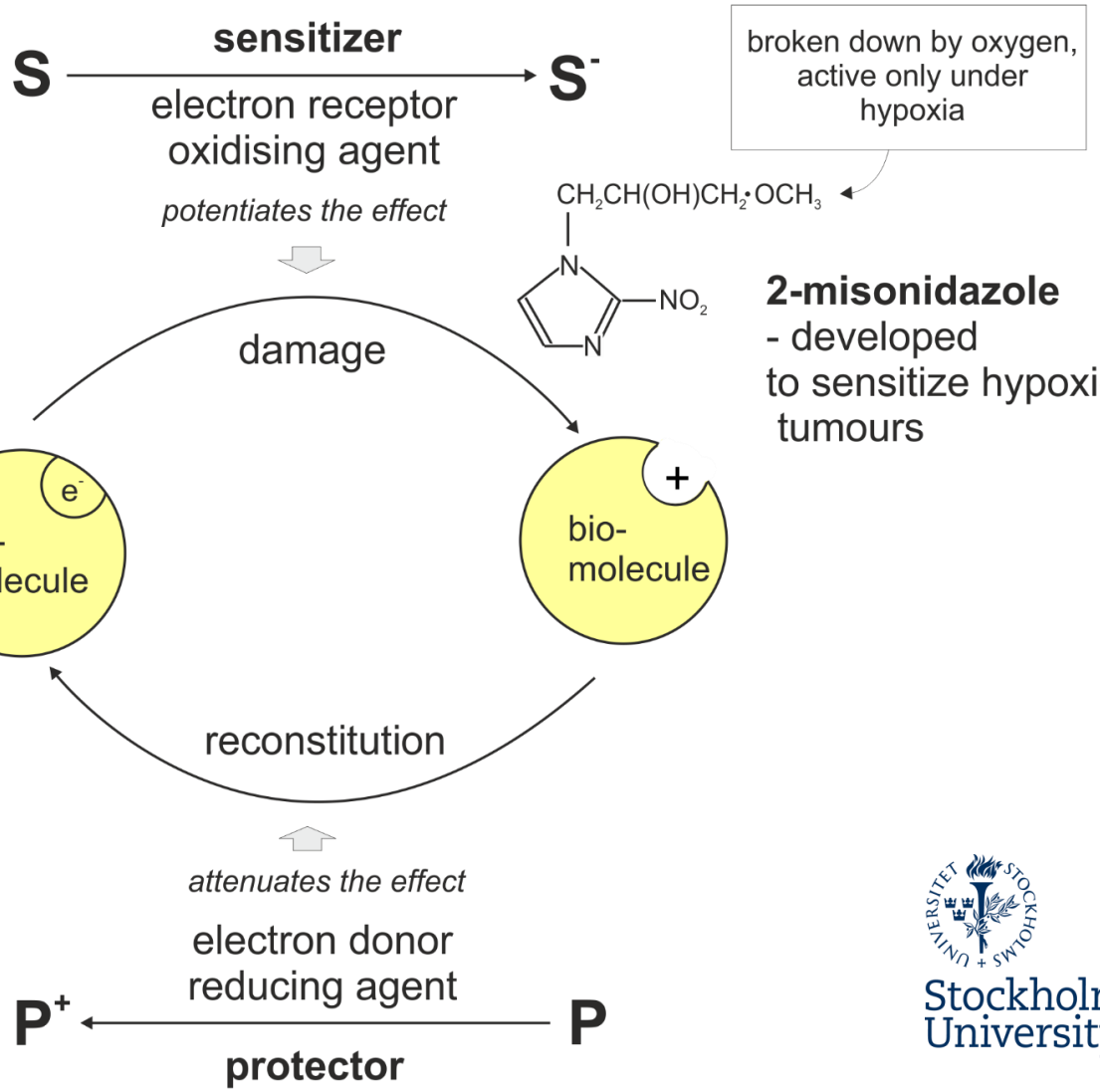
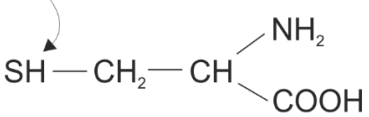
Radiosensitisers and radioprotectors

Radioprotectors and radiosensitisers act on the indirect effect of radiation



cysteine - a precursor to glutathione

the nucleophylic SH (thiol) group



glutathione
amifostine

developed as radioprotector

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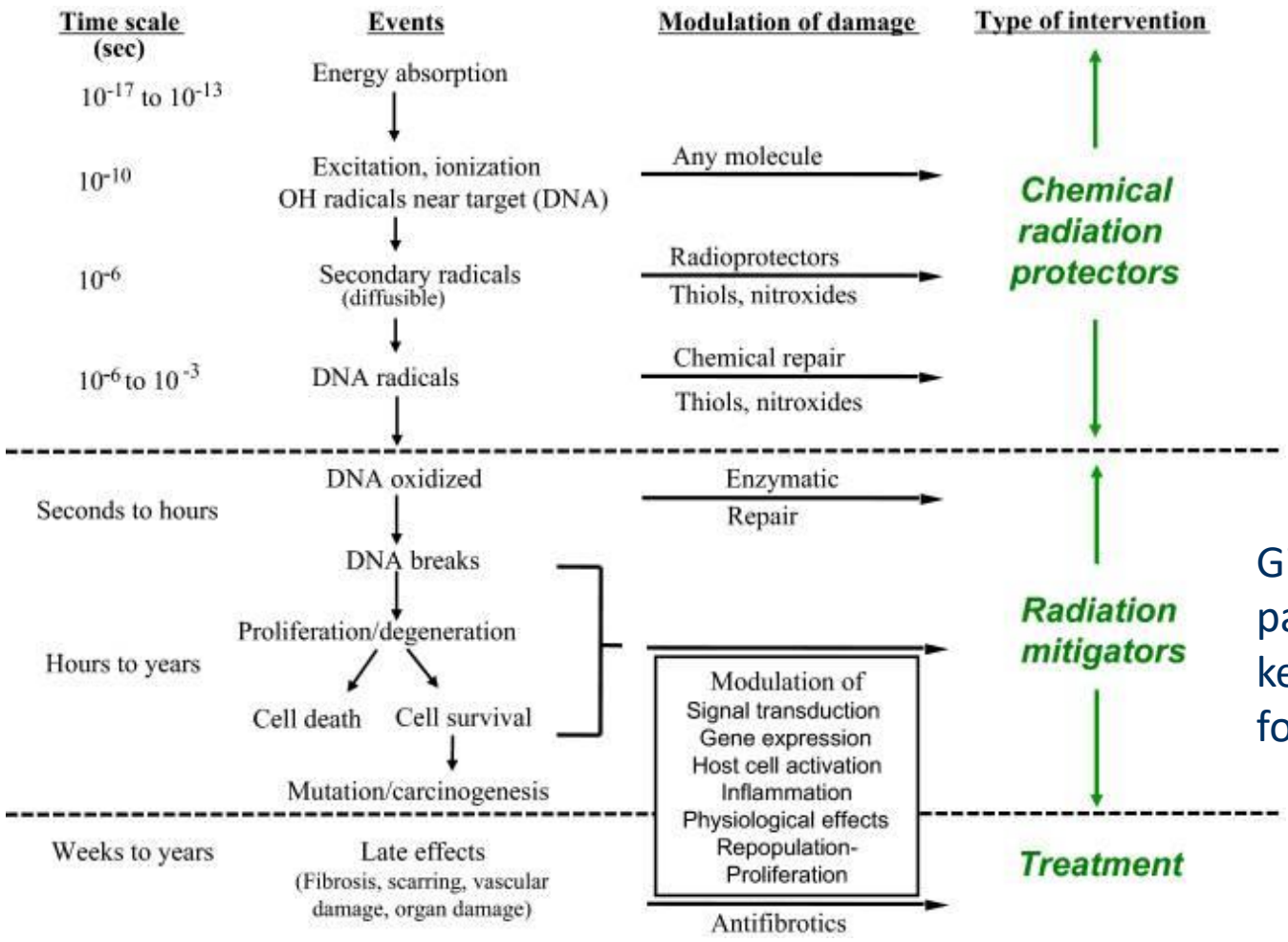
Radioprotectors and mitigators



Given prior to radiotherapy, ex. amifostine

Given after radiotherapy, ex. palifermin, a recombinant keratinocyte growth factor for mitigation of mucositis

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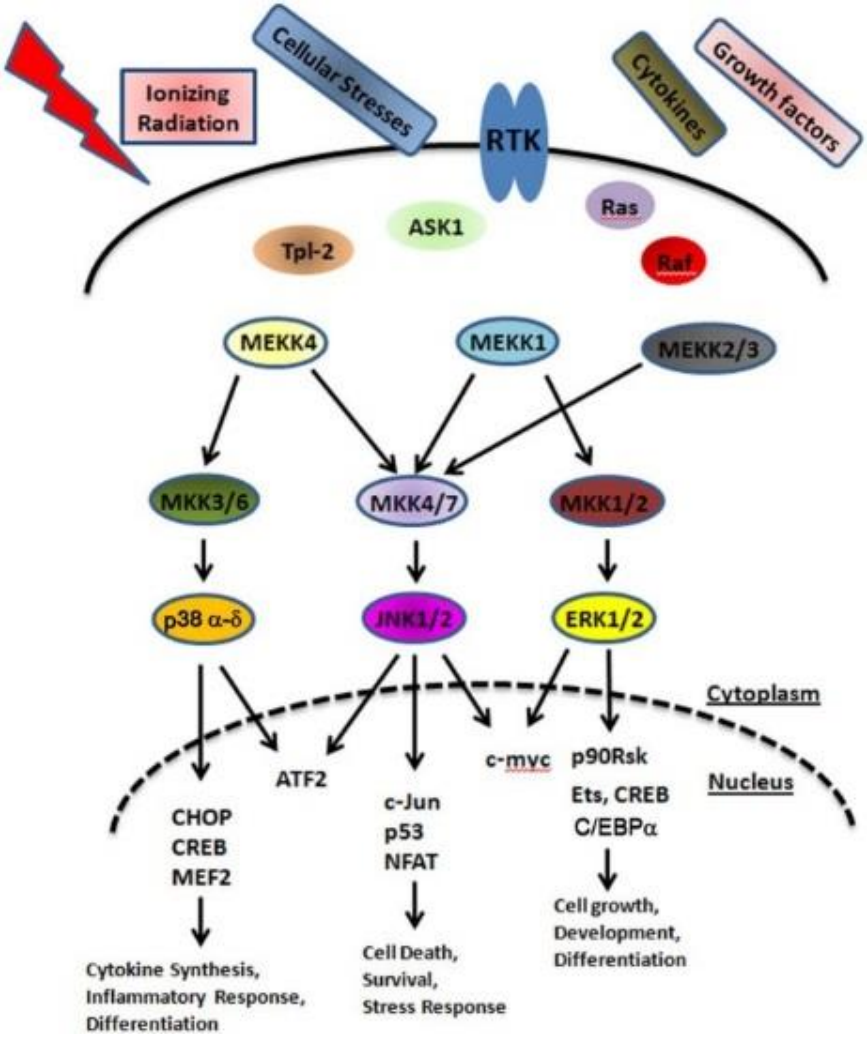
Citrin et al. The Oncologist, 2010

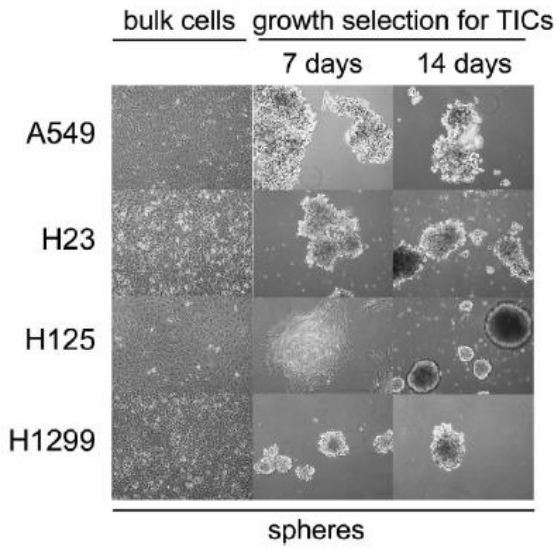
Other types of radiosensitizers

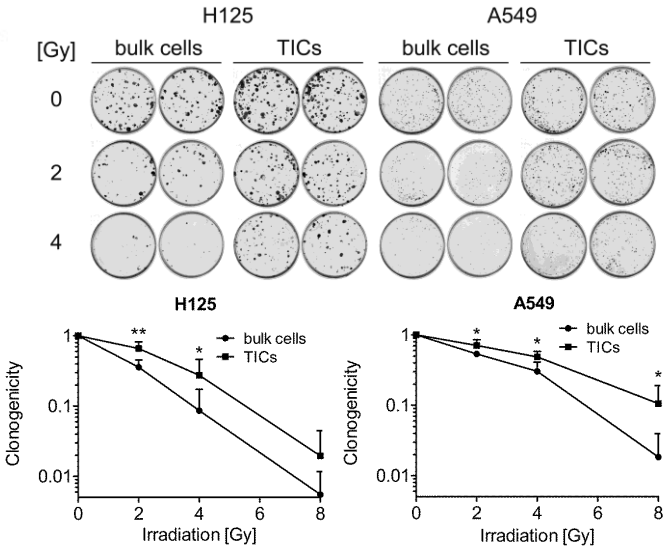
- Chemotherapeutics
  - Cisplatin or other platinum analogs: Inhibits DNA repair by crosslinking strands
  - Gemcitabine: Depletion of dATP pools, S-phase blockage, lowered threshold for radiation-induced apoptosis Lawrence et al. Oncology 1999
- Metformin (hyperglycemia/diabetes drug)
  - Impairment of oxidative phosphorylation Van Gisbergen et al. Mutat Res Rev Mutat Res. 2015
- Receptor tyrosine kinase (RTK) inhibitors
  - MEK inhibitors
- Chromatin modifiers
  - Histone deacetylase inhibitors opens up the chromatin structure

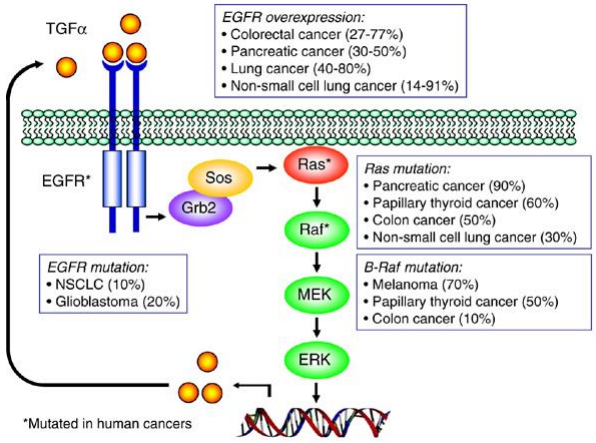


Munshi et al. Genes Cancer. 2013

Example: MEK inhibition + gamma radiation targets non-small cell lung cancer stem cells

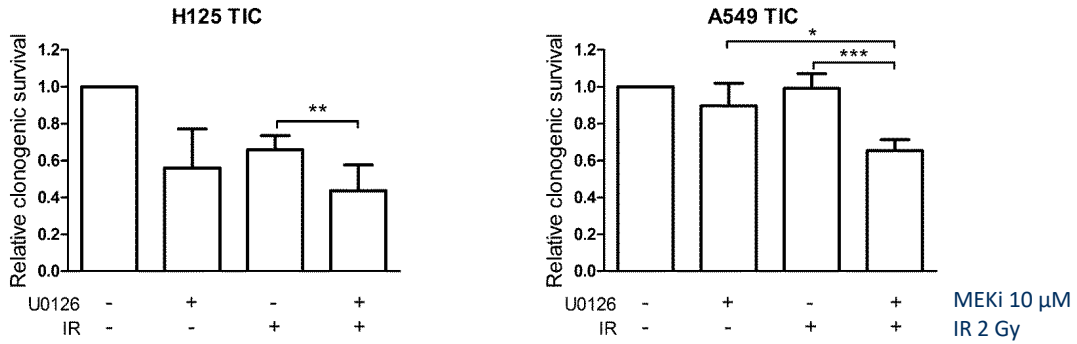






Tumour initiating cells (TICs, or CSC) are enriched from non-small cell lung cancer (NSCLC) cell lines by growth for 10 days in non-adherent conditions, in serum-free media supplemented with growth factors, hormones and heparin

Lundholm et al. Cell Death Dis. 2013



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Lundholm L et al., IJRB 2014

Factors influencing cellular radiosensitivity

- Physical factors
  - Dose, radiation quality, dose rate, fractionation, temperature
- Chemical factors
  - Oxygen, radiosensitisers, radioprotectors
- **Biological factors**
  - Organism level: Whole/partial body exposure, age, inherited genetic disorders, inflammatory state/immune response/infections/microbiome
  - **Cellular level: Cell cycle stage, stem cell/differentiated cell type, chromatin conformation**
- Technical factors
  - Accuracy of radiotherapy delivery

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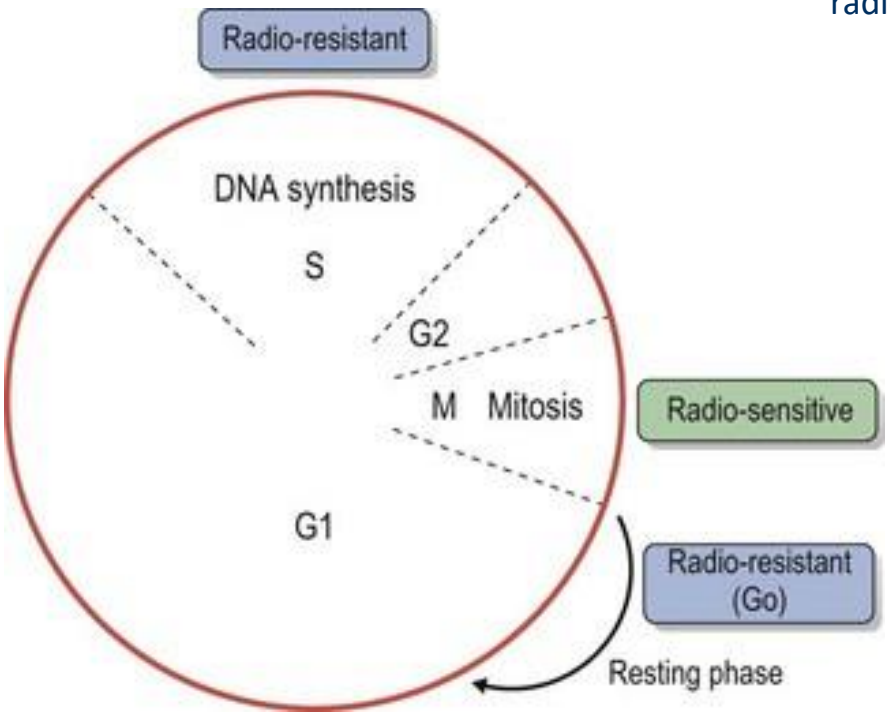
Cell cycle stage

Highest > lowest sensitivity

M > G2 > G1 > early S > late S

- No time for adequate repair before chromosome segregation takes place

- An elevated amount of DNA synthesis and repair enzymes
- Elevations in the intracellular levels of glutathione (a free radical scavenger)



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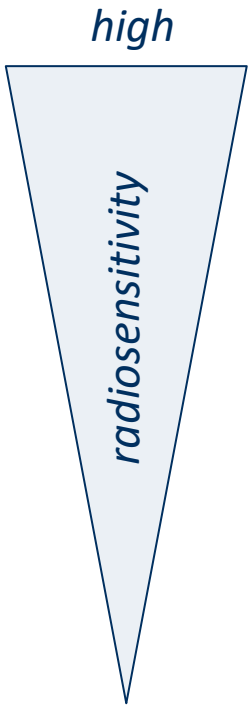
Stem/differentiated cell type

The most radiosensitive cells are those which:

- Have a high division rate — Little time for repair of damage
- Have a high metabolic rate — A lot of energy to undergo apoptosis
- Are of non-specialized type — High proliferation activity
- Are well nourished — A lot of energy to undergo apoptosis



Lymphoid organs, bone marrow, blood, testes, ovaries, intestines

Skin and other organs with epithelial cell lining (cornea, oral cavity, esophagus, rectum, vagina, uterine cervix, urinary bladder, ureters)

Fine vasculature, growing cartilage, growing bone

Mature cartilage or bone, salivary glands, respiratory organs, kidneys, liver, pancreas, thyroid, adrenal and pituitary glands

Muscle, brain, spinal cord

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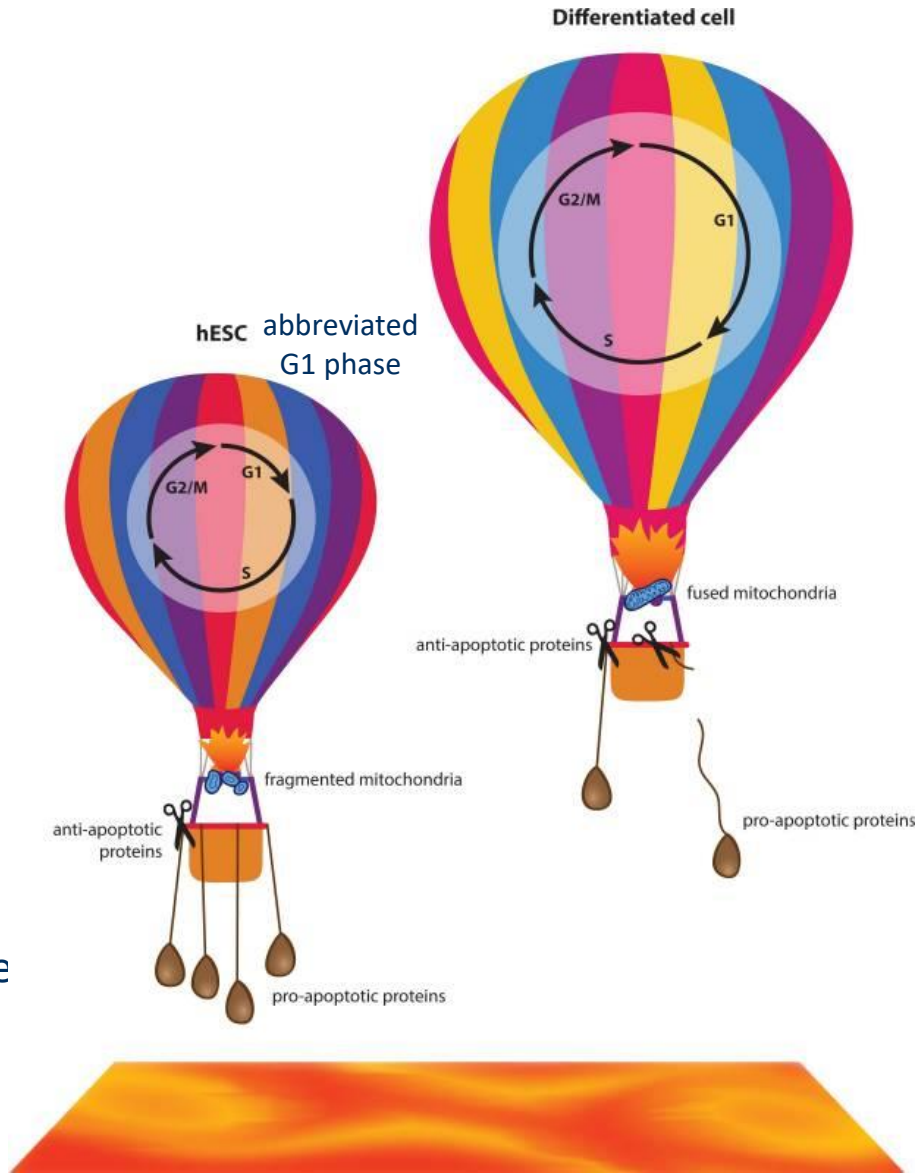
Rubin, P. and Casarett. G. W. Cancer. 1968

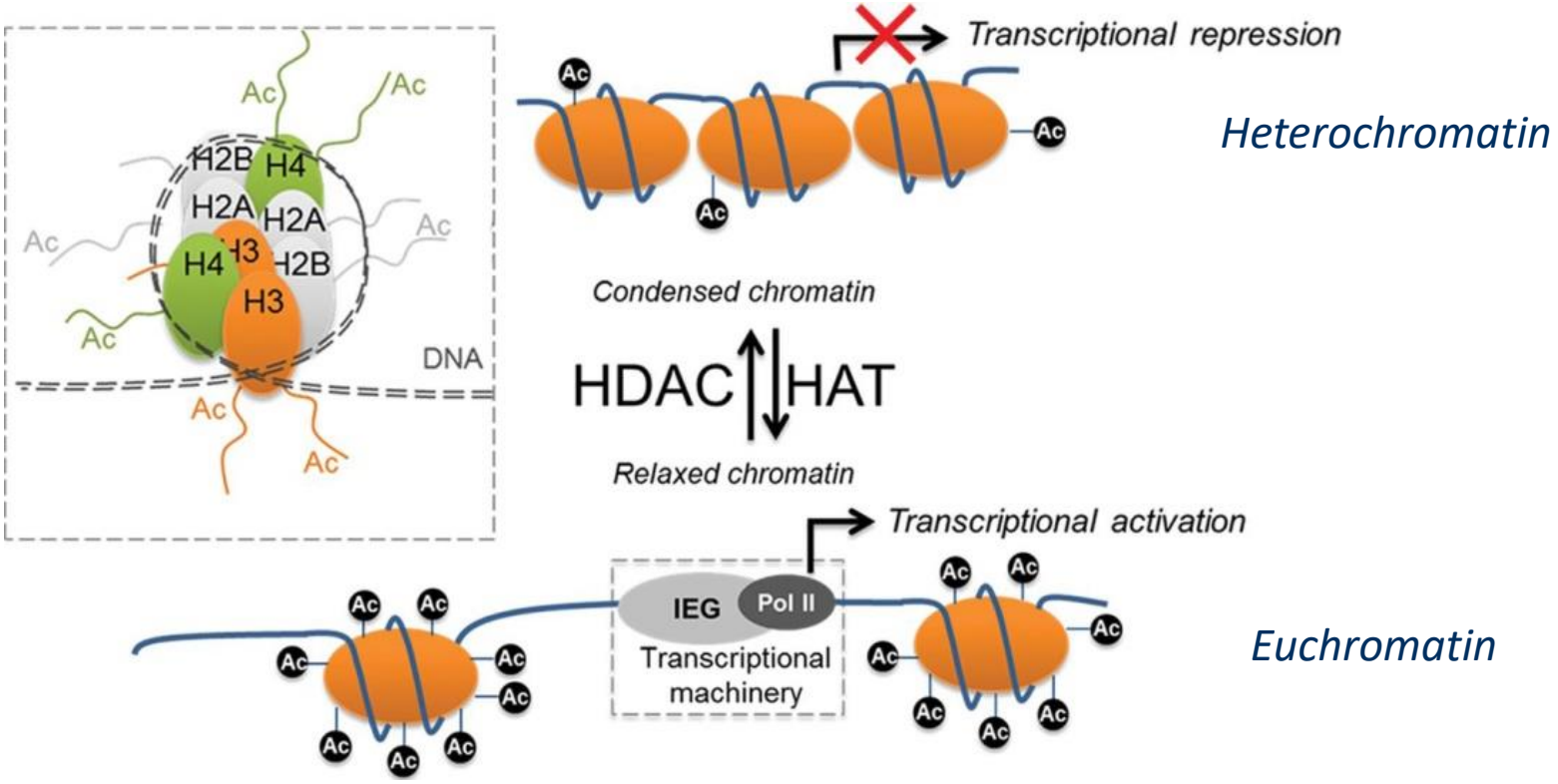
Radiosensitivity of stem vs differentiated cells

- **Embryonic stem cells (ESC)**
  - Very radiosensitive
  - Give rise to all the tissues in the body, therefore prone to undergo apoptosis after damage to avoid compromising the genomic integrity of the population
- **Adult stem cells**
  - Variable radiosensitivity, due to dual roles:
    - More resistant to cell death, possibly to prevent uncontrolled apoptosis that might compromise tissue and organ structure
    - Sensitive enough to avoid genomic instability in progeny if damage-induced mutations are not properly repaired
- **Differentiated cells**
  - Relatively radioresistant, longer G1 phase
  - Transcriptional repression of ATM may contribute (differentiated astrocytes compared to neural stem cells) Schneider et al. Cell Death and Differentiation (2012) 19, 582–591



Liu et al. Trends Cell Biol. 2014

Chromatin conformation



Immediate early genes (IEGs)

Whittle et al. Biochemical Society Transactions 2014

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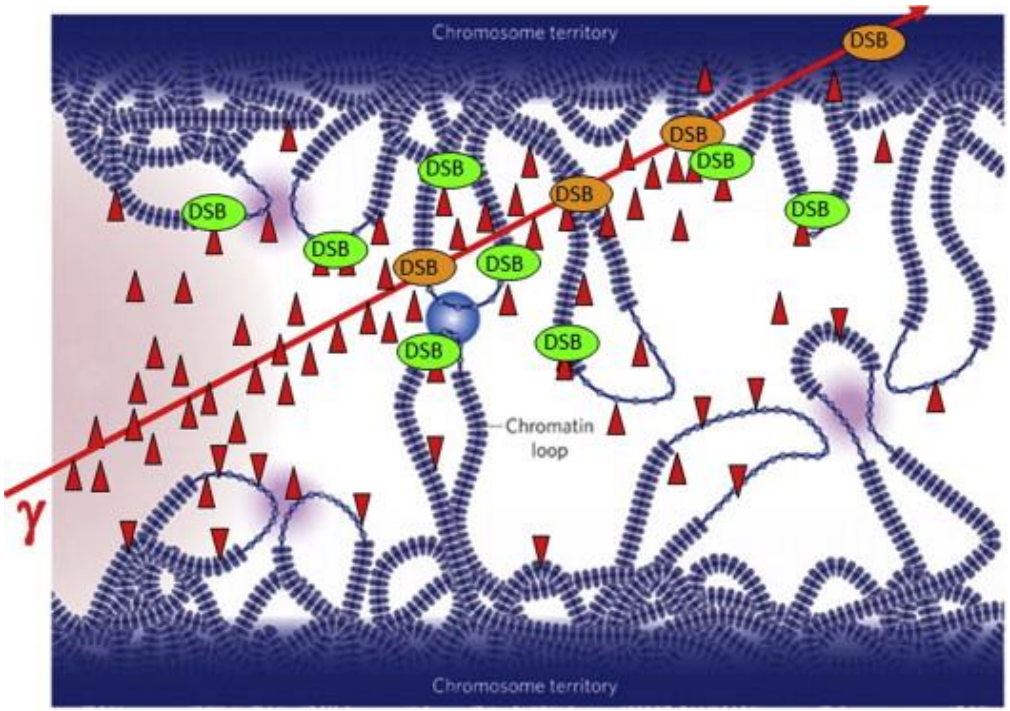
Decondensed chromatin/euchromatin is more sensitive to low LET radiation (photons)

- Induction of DSBs by low LET radiation from:
  - Direct effects (30%)
  - Indirect effects (70%), mediated by reactive free radicals produced especially by the water radiolysis
- More radicals are produced in decondensed chromatin due to its high hydration
  - The radicals are short-lived and damage DNA close to their sites of induction
- Dense heterochromatin composition (compaction, and a larger amount of proteins) shields the DNA better from the harmful radicals



The background image (chromatin) is taken from P Fraser & W Bickmore (2007) Nature 447, 413-417, http://www.nature.com/nature/journal/v447/n7143/images/nature05916-f2.2.jpg

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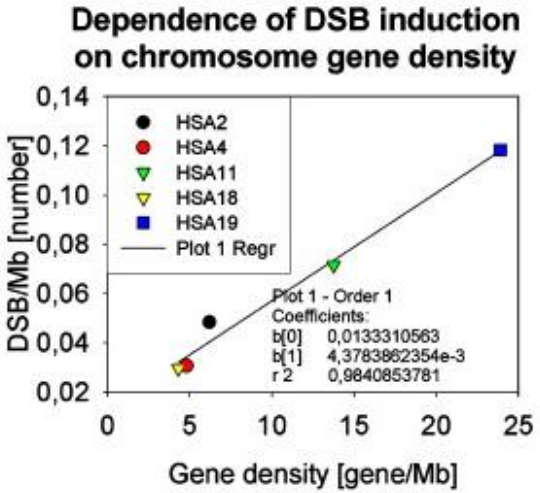
Falk et al. Applied Radiation and Isotopes, 2014

Fewer DSBs in chromosomes with lower gene density

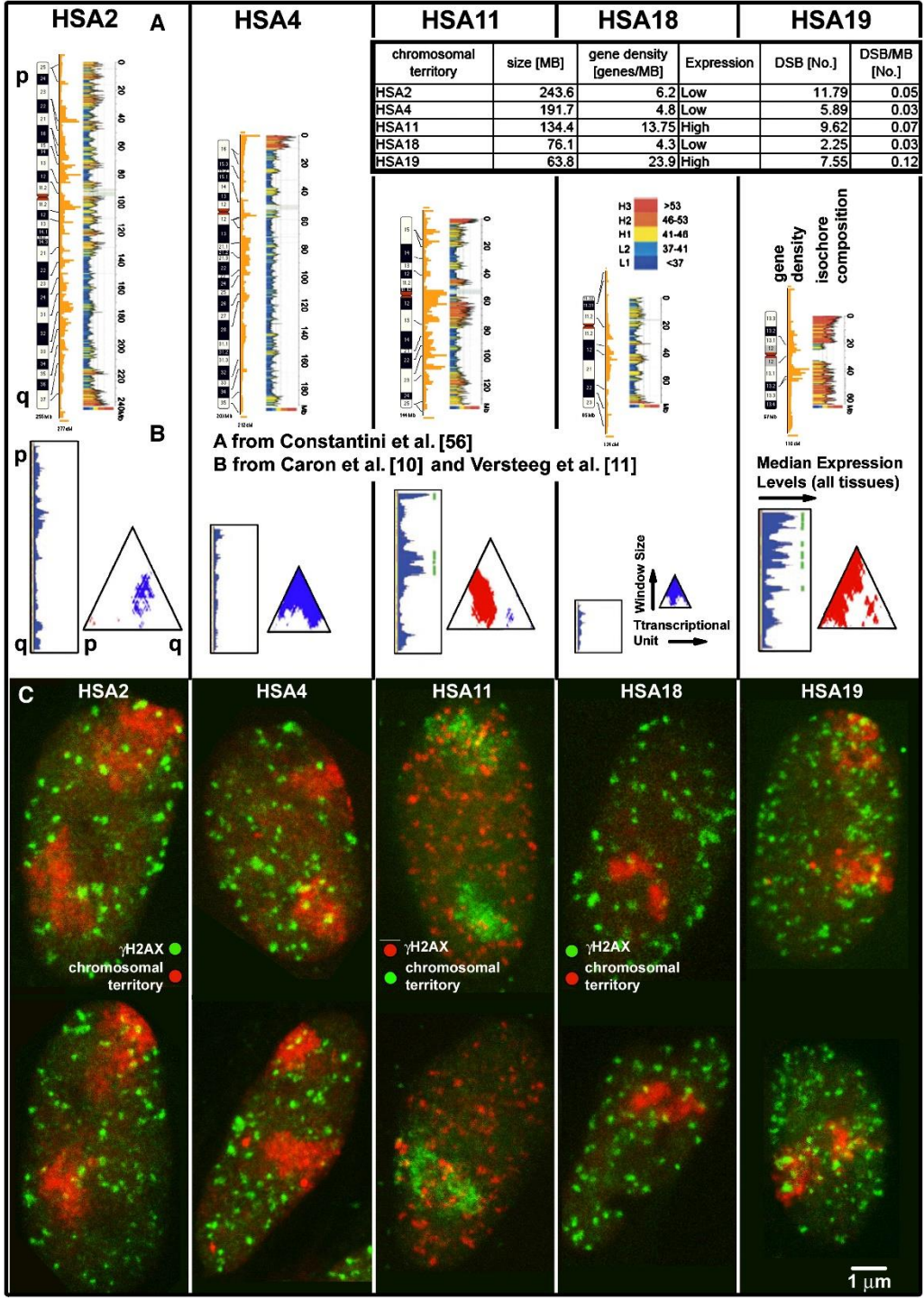
- Fewer gH2AX foci (DSBs)/megabase DNA for chr 2, 4 and 18 than for chr 11 and 19



Fibroblast nuclei with simultaneously visualized (ImmunoFISH) territories of specific chromosomes (red; green for HSA11) and induced γH2AX foci (green; red for HSA11).

To explore the role of chromatin structure in radiosensitivity of cells, the osmolarity of the medium was changed.

Falk et al. BBA 2008

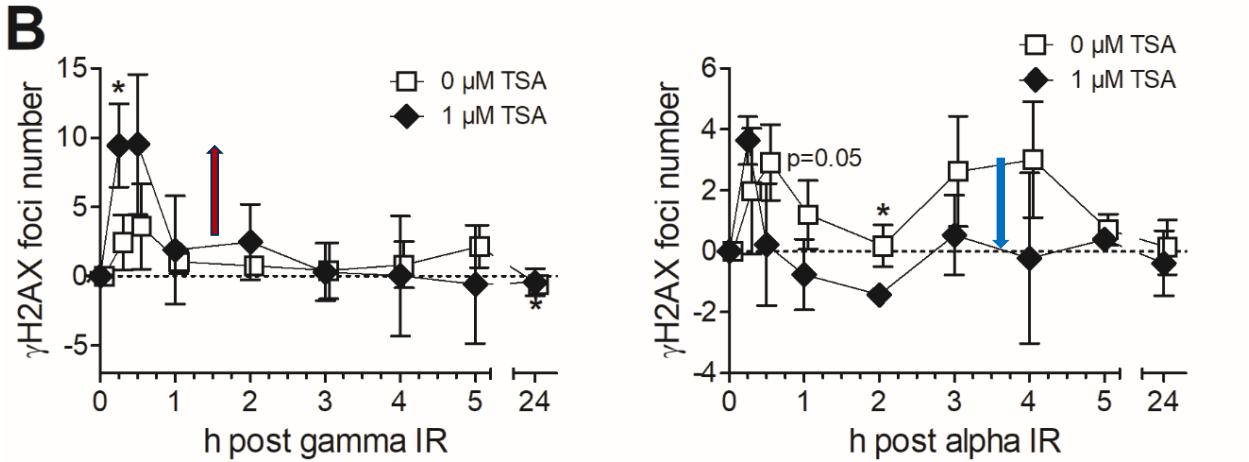


chromosomal territory	size [MB]	gene density [genes/MB]	Expression	DSB [No.]	DSB/MB [No.]
HSA2	243.6	6.2	Low	11.79	0.05
HSA4	191.7	4.8	Low	5.89	0.03
HSA11	134.4	13.75	High	9.62	0.07
HSA18	76.1	4.3	Low	2.25	0.03
HSA19	63.8	23.9	High	7.55	0.12

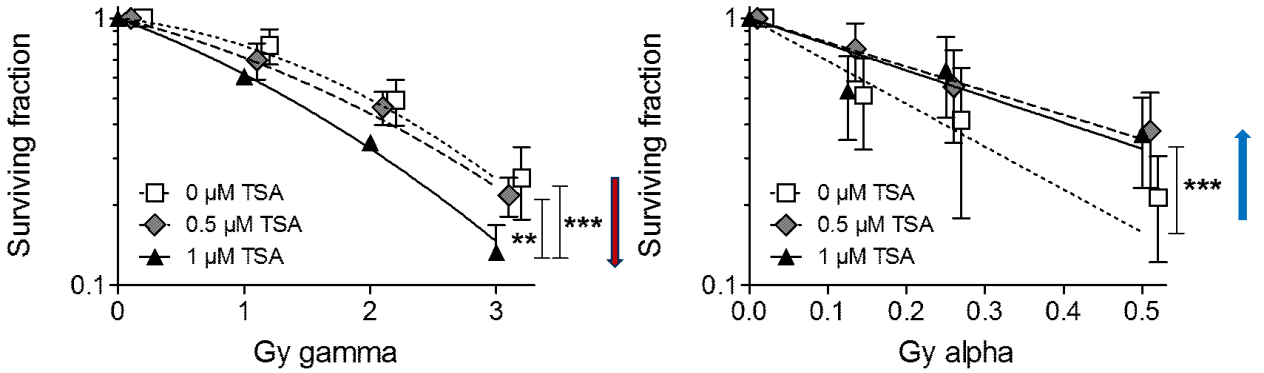
Example: Is the chromatin protective also using high LET alpha radiation?

When opening chromatin before:

- High LET damage - Improved DNA repair appears to be most important
- Low LET damage - Increased DNA damage is dominant



Histone deacetylase inhibitor: Trichostatin A (TSA)



Chromatin opening using a histone deacetylase inhibitor (HDACi) gives opposite effects after gamma and alpha radiation in breast cancer MDA-MB-231 cells

Stockholm University

Svetličič M et al. Cells 2020

Conclusions

A number of factors influence the cellular radiosensitivity:

- Radiation quality, dose, dose rate and fractionation schedule for induced DNA damage
- DNA repair capacity
- Choice of cell death pathway - partially dependent on cell type
  - Apoptosis, senescence, etc
- Oxygen levels, presence of antioxidants
- Stemcellness
- Chromatin state

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