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Individual response to ionizing radiation: radiosensitivity, radiosusceptibility and radiodegeneration

Michel Bourguignon, MD, PhD

Professeur de Biophysique et médecine nucléaire

Faculté de médecine Simone Veil

Université Paris Saclay / UVSQ

INSERM UA8 « Radiations: Défense, Santé, Environnement »

pr.michel.bourguignon@gmail.com

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Individual response to ionizing radiation

We are not equal vis-à-vis IR : an old idea, a forgotten evidence, an economic and societal issue

- December 1895 : Roentgen discovers X rays
- February 1896: Becquerel discovers radioactivity

- 1896 : first radiotherapy by Victor Despeignes
- 1901 : first description by Pierre Curie of deleterious cutaneous effects of radium rays
- 1902 : first description by Frieben of radio-induced cancer
- 1911 : first description by Léon Bouchacourt of a difference of sensitivity to X rays, i.e., skin response after high dose IR

N.Foray, C.Colin, M.Bourguignon. 100 years of individual radiosensitivity: how we have forgotten the evidence.Radiology, 2012, 264 :627-31

1911 : First publication about individual radiosensitivity

Dr. Léon Bouchacourt
(1865-1949)

To be peeled by… X-rays…

M. L. BOUCHACOURT,

Chef du Service de Radiologie de l'Hôpital Dubois.

SUR LA DIFFÉRENCE DE SENSIBILITÉ AUX RAYONS DE RÖNTGEN DE LA PEAU DES DIFFÉRENTS SUJETS, ET, SUR LE MÊME SUJET, DES DIFFÉRENTES RÉGIONS DU CORPS.

QUE DOIT-ON PENSER DU TRAITEMENT RADIOTHÉRAPIQUE DE L'HYPERTRICHOSE ET DE L'HYPERHIDROSE?

…" About the differences in sensitivity to Roentgen rays for different individuals, and for a given individual, for different body locations"…

radiosensitivity = tissue reactions

Foray et al. Med Sci. 2013;29(4):397-403

Reviews on individual response to ionizing radiation (1)

Historical reviews

- C Alapetite, JM Cosset, M Bourguignon, R. Masse. Genetic susceptibility to radiations. Which impact on medical practice ? *Quart J Nucl Med* 2000, 44 : 347-354
- Bourguignon MH, Gisone PA, Perez MR, Michelin S, Dubner D, Giorgio MD, Carosella ED. Genetic and epigenetic features in radiation sensitivity Part I: Cell signalling in radiation response. *Eur J Nucl Med Mol Imaging.* 2005 ; 32(2):229-46.
- Bourguignon MH, Gisone PA, Perez MR, Michelin S, Dubner D, Giorgio MD, Carosella ED. Genetic and epigenetic features in radiation sensitivity Part II: implications for clinical practice and radiation protection. *Eur J Nucl Med Mol Imaging*. 2005 ; 32(2):351-68.
- Human radiosensitivity, HPA 2013
- Foray, Bourguignon,Hamada. Individual response to ionizing radiation. *Mutation Research* 770 (2016)

Reviews on individual response to ionizing radiation (2)

Three new and large literature reviews (European consortium MELODI)

- Seibold, Auvinen, Averbeck, Bourguignon et al., Clinical and epidemiological observations on individual radiation sensitivity and susceptibility. *International Journal of Radiation Biology 2019*
- Averbeck D et al. Establishing the mechanisms affecting the individual response to ionising radiation, including the contribution of any genetic component. *International Journal of Radiation Biology 2019*
- Gomolka, Blyth, Bourguignon et al. Potential screening assays for individual radiation sensitivity and susceptibility and their current validation state. *International Journal of Radiation Biology 2019*

- Kalman and Oughton. Ethical considerations related to radiosensitivity and radiosusceptibility. *International Journal of Radiation Biology 2019*

Creation of ICRP TG111 : (Nov 2018) –Factors Governing the Individual Response of Humans to Ionizing Radiation

Individual response to ionizing radiation

A fruitful clinical approach

Three types of patients/situations

- Patients with radiation-induced adverse tissue events attributable to cell deaths and loss of clonogenicity, i.e., complications and undesirable side effects of radiotherapy (with no error in dose delivery) which affect the quality of life and are not cancer effects = radiosensitivity
- Patients with cancer proneness and especially after exposures to ionizing radiation, linked to survival of DNA transformed cells and genomic instability = radiosusceptibility
- Patients with late tissue degeneration after exposures to ionizing radiation, e.g., cataracts or cardiovascular effects, linked to cell and tissue ageing = radiodegeneration

Semantic clarification

- Radiosensitive persons are also radiosusceptible but one can be radiosusceptible (and cancer prone) without being radiosensitive (e.g., Li Fraumeni p53)
- Children are more radiosensitive than adults ! When do they stop ?
- Women are more radiosensitive means that they are more cancer prone, i.e., radiosusceptible

- Foray N, Bourguignon M and Hamada N. Individual response to ionizing radiation. Mutation Research 770 (2016) : 369–386
- Britel M, Bourguignon m, Foray N. The use of the term 'radiosensitivity' through history of radiation: from clarity to confusion. Int J Radiat Biol 94(2018) : 503-512

Individual response to ionizing radiation

A public health issue

Up to 20% of the population is concerned

- But a continuum between normal and highly pathological response
- Large population exposed to low doses ionizing radiation : medical (repeated exposures), radon, cosmic (planes), dismantling, nuclear accidents…
- The risks are higher than in normal population (e.g., family risk of breast cancer with no identified gene) but not taken into account by ICRP

Medical exposures are the most important of all exposures and steadily increase = focus on patients

- Radiotherapy of 50% of cancers with 80% cure but 8 % of radio-induced cancers
- Diagnostic CT and interventional radiology (58% of medical doses)

Combined risks of exposures to low doses of genotoxic compounds (chemicals…) = exposome with potentially supra-additive effects

1981 : First correlations with individual radiosensitivity

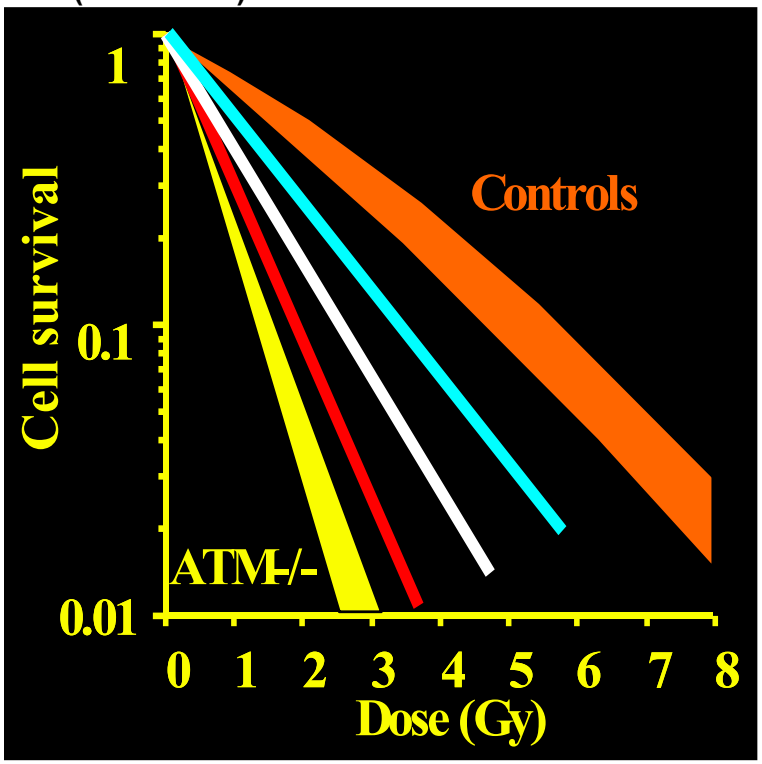
Dr. EP Malaise
(1930-2013)

Human clinical radiosensitivity = in vitro surviving fraction at 2 Gy

- Survival curves never cross : intrinsic radiosensitivity

- There is a continuum in radiation responses

- Quantitative correlation between survival fraction at 2 Gy (SF2) and local tumor control



Fertil and Malaise, Int J Radiat Biol Oncol Phys, 1981, 7(5):621-9
Deschavanne and Fertil, Int J Radiat Biol Oncol Phys, 1996, 34(1):251-66

The genetic syndromes associated with radiosensitivity : an obvious link to DNA DSB repair (70%)

but there are exceptions (30%)

SYNDROMES	MUTATED GENE	SF2
Ataxia telangiectasia (classical homoz.)	ATM	1-5
Syndrome Ligase IV	LIG IV	2-6
Nijmegen syndrome	NBS1	5-9
➡ Progeria	Lamin A	8-19
Ataxia telangiectasia (variant homoz.)	ATM	10-15
➡ Usher's syndrome	USH	15-20
Cockayne's syndrome	CS	15-30
Xeroderma Pigmentosum	XP	15-30
AT-Like Disorder	MRE11	15-40
➡ Huntington Chorea	IT15	18-30
➡ Gardner's syndrome	APC	20-30
Turcot's syndrome	hMSH2	20-30
Fanconi anemia and BRCA2 mutations	FANC	20-40
BRCA1 mutations	BRCA1	20-40
Artemis mutations	Artemis	20-40

1 to 40 x

Deschavanne and Fertil, Int J Radiat Biol Oncol Phys, 1996, 34(1):251-66

The major human syndromes associated with radiosensitivity and/or radiosusceptibility

Table 1
The major human syndromes associated with radiosensitivity and/or radiosusceptibility[a].

Syndromes	Mutated Genes	Major defective mechanism	Cancer predisposition	Clinical sensitivity to IR[b]	SF2 of fibroblastic and normal cell strains [a,b]
Ataxia telangiectasia	ATM homozygous mutations	DSB signaling and repair	Leukemia, Lymphoma	+++	1–5
Ligase IV	Lig IV homozygous mutations	NHEJ	Leukemia, Lymphoma	+++	2–6
Nijmegen	NBS1 homozygous mutations	DSB signaling and repair	Leukemia, Lymphoma	+++	5–9
Hutchinson-Gilford (progeria infantum)	Lamin A homozygous mutations	Nuclear membrane	No	+++	8–19
Bruton's disease (agammaglobulinemia)	BTK homozygous mutations	V(D)J recombination	No	+++	10
Hypo-gammaglobulinemia	Lig I	NER	No	+++	11
Glutathione synthetase deficiency	GSS	Glutathione cycle	No	+++	14
ICF syndrome	DNMT3B	DNA methylation DSB signalling and repair	No	+++	14
Huntington's disease	IT15	DNA methylation DSB signaling and repair	No	++	19
Neurofibromatosis type I (Von Recklinghausen)	NF1	DSB signaling and repair	Central and peripheral nervous system tumors	++	15–20
Tuberous sclerosis	TSC genes	DSB signaling and repair	Central and peripheral nervous system tumors	++	24
Cockayne syndrome	CS genes	NER/TCR	Skin cancers but not for all mutations genes	0/++	15–30
Xeroderma pigmentosum	XP genes	NER/TCR	Skin cancer	0/++	15–30
AT like disorders	MRE11	DSB signaling and repair	?	0/++	15–40
Fanconi anemia	FANC gene	HR	Leukemia, squamous cell carcinoma, breast cancer	0/++	15–40
$AT^{+/-}$	ATM heterozygous mutations	DSB signaling and repair	High risk of breast cancer	0/++	20–30
Li-Fraumeni syndrome	p53	DSB signaling and repair Cell cycle regulation Apoptosis regulation	Different types of cancer: breast, brain, leukemia, sarcoma	0/+	20–30
Turcot and Gardner syndromes	APC genes	Cell adhesion	Mainly colorectal cancer	0/+	20–30
Severe combined immunodeficiency	Artemis Cernunos/XLF	V(D)J recombination NHEJ	Lymphoma	0/+	20–40
Hereditary breast/ovary cancer	BRCA2	HR	Breast/ovary cancer	0/+	20–40
Nevoid basal cell carcinoma (Gorlin) syndrome	PTCH1	Embryonic structures Proliferation regulation Membrane trafficking	Non-melanoma skin cancer	0/+	20–50
Hereditary retinoblastoma cancer	RB1	DSB signaling and repair Cell cycle regulation Apoptosis regulation	Retinoblastoma, sarcoma, melanoma, lung cancer, breast cancer	0/+	30–50
Hereditary breast/ovary cancer	BRCA1	HR	Breast/ovary cancer	0/+	30–50
Bloom's syndrome	BLM RecQ	HR/TLS	leukemia, lymphoma	0/+	30–50
Rothmund-Thomson syndrome	RecQL4	HR/TLS	osteosarcoma	0/+	30–50
Werner syndrome	WRN RecQ	HR/TLS		0/+	30–50
Hereditary non polypoid colorectal cancer (Lynch syndrome)	hMLH1, hMSH2/6, hPMS2	MMR	Colorectal cancer	0/+	30–50
Radioresistance	–	–	No	0	50–70 [a]

Foray N, Bourguignon M and Hamada N. Individual response to ionizing radiation. Mutation Research 770 (2016) : 369–386

- 27 syndromes with a genetic trait in this list
- All together > 5% of the population

(ATM+/- = 1.5 %)

- Many other patients without known genetic trait exhibit clinical radiosensitivity
- Up to 20% of the population ?

Existing cohorts and biobanks suitable for radiosensitivity research

Dedicated radiosensitivity cohorts composed of patients referred to radiation biology studies for clinical radiosensitivity

- COPERNIC (France)
- RILA (France + various countries)
- RTOG/EORTC trials (various countries)

Other oncology cohorts

- REQUITE (EU)

Other cohorts and biobanks

- Biobank of Eastern Finland
- CONSTANCES (France)
- EPIC (IARC)
- NAKO (Germany)
- UK Biobank

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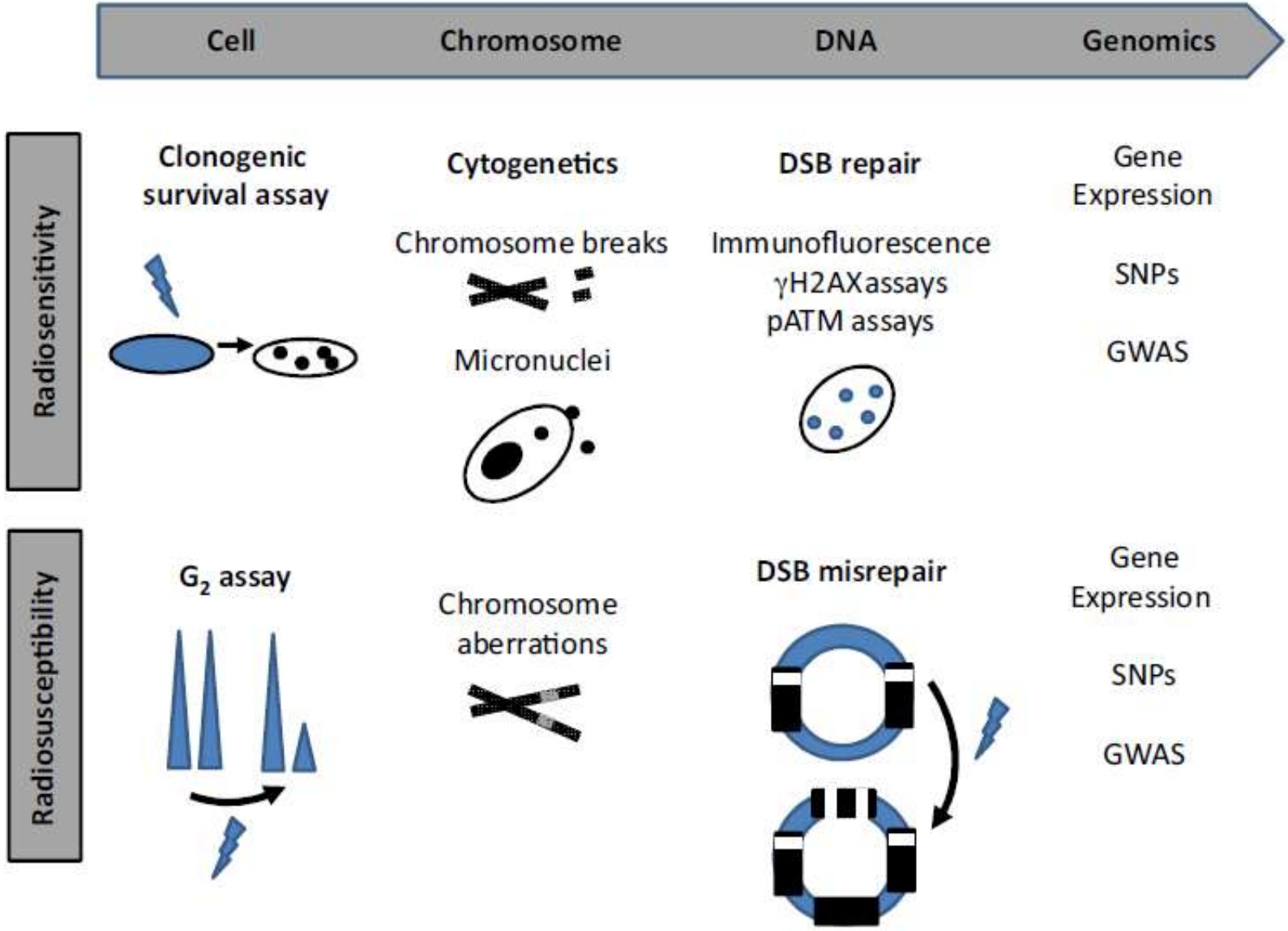
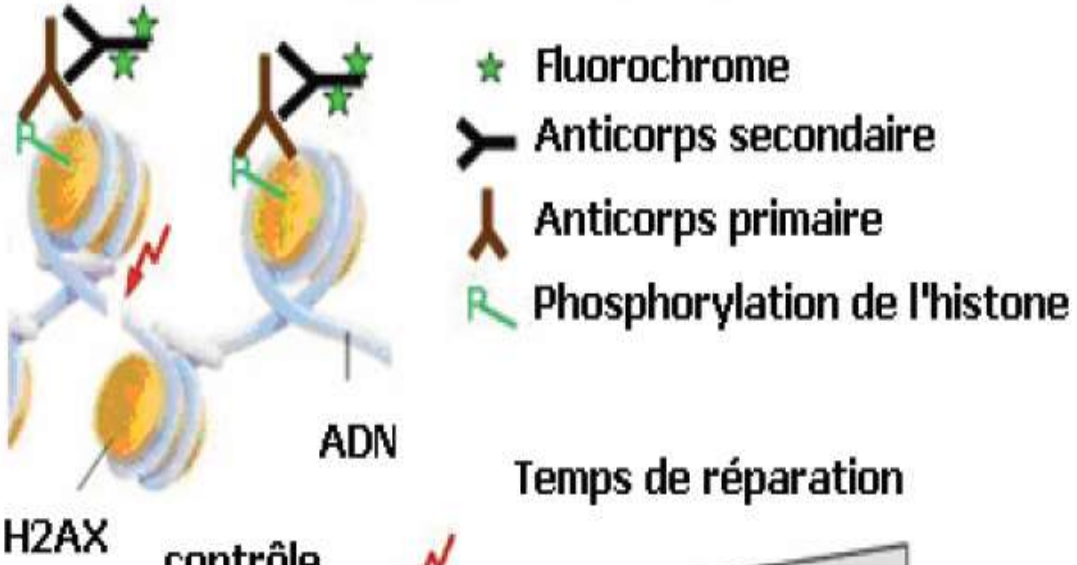
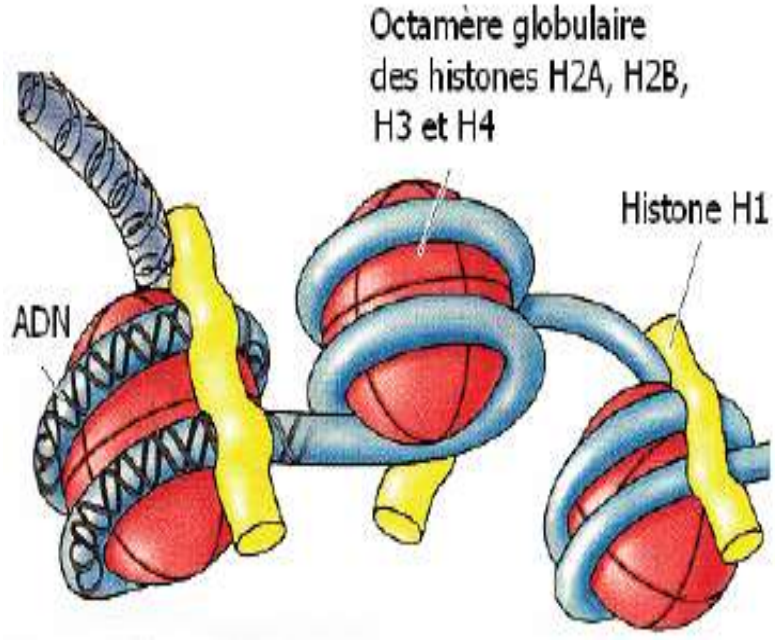
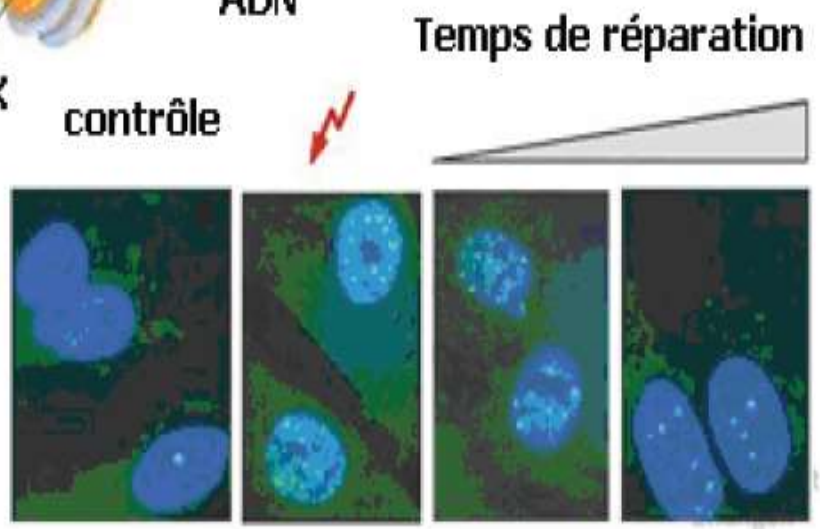
Major assays for evaluation of radiosensitivity and radiosusceptibility (not suitable for screening in routine clinical practice)



Immunofluorescence: a powerful technique of investigation





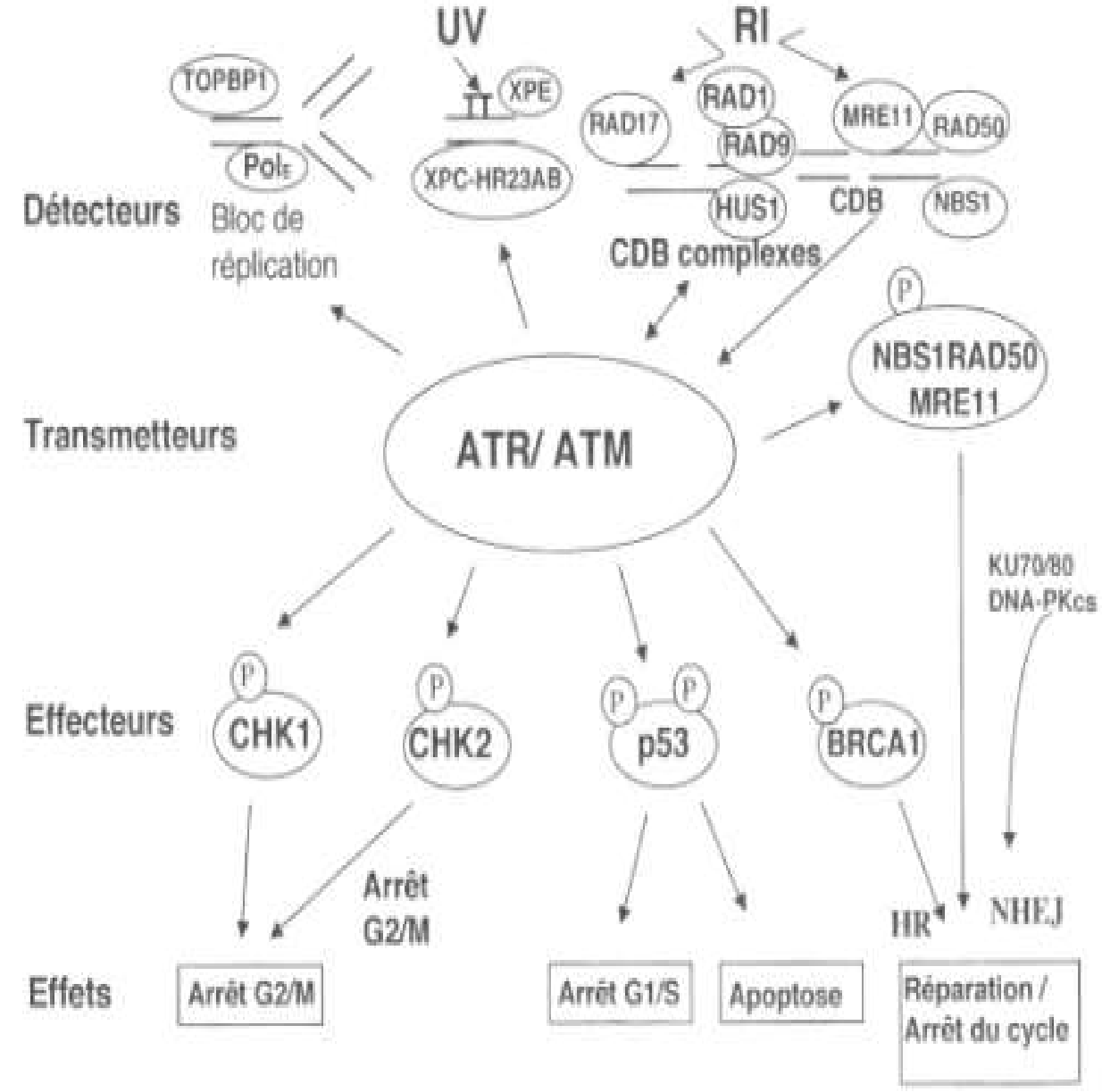


Blue coloration of chromatin with DAPI

Visualisation of radioinduced damages with immunofluorescent γH2AX foci

Immunofluorescence: a powerful technique of investigation

- Identification and quantification of DNA DSBs at the dose de 1 mGy by immunofluorescence anti-histones γH2-AX *(Rothkamm & Löbrich 2003) :*
  - gain in sensitivity of 100 : threshold of 1 mGy
  - The effects of one single radiologic examination (e.g., one mammography) can be visualized and quantified
- Identification of poorly repaired DNA DSBs by immunofluorescence MRE11
- Other markers of signalization and repair pathways of DNA lesions (ATM, 53BP1, …) can be studied by immunofluorescence

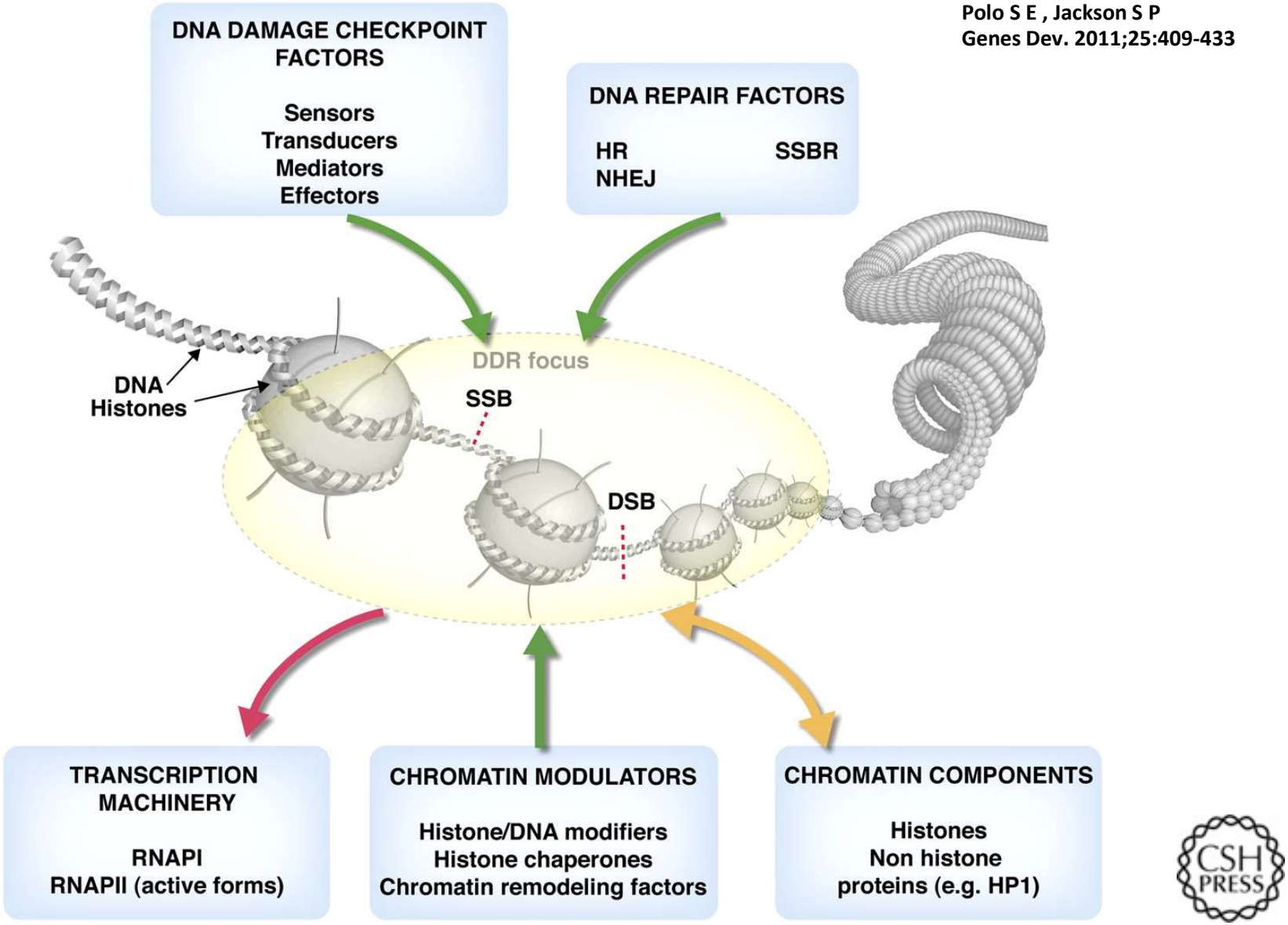


Cascade of signalization after DNA damage

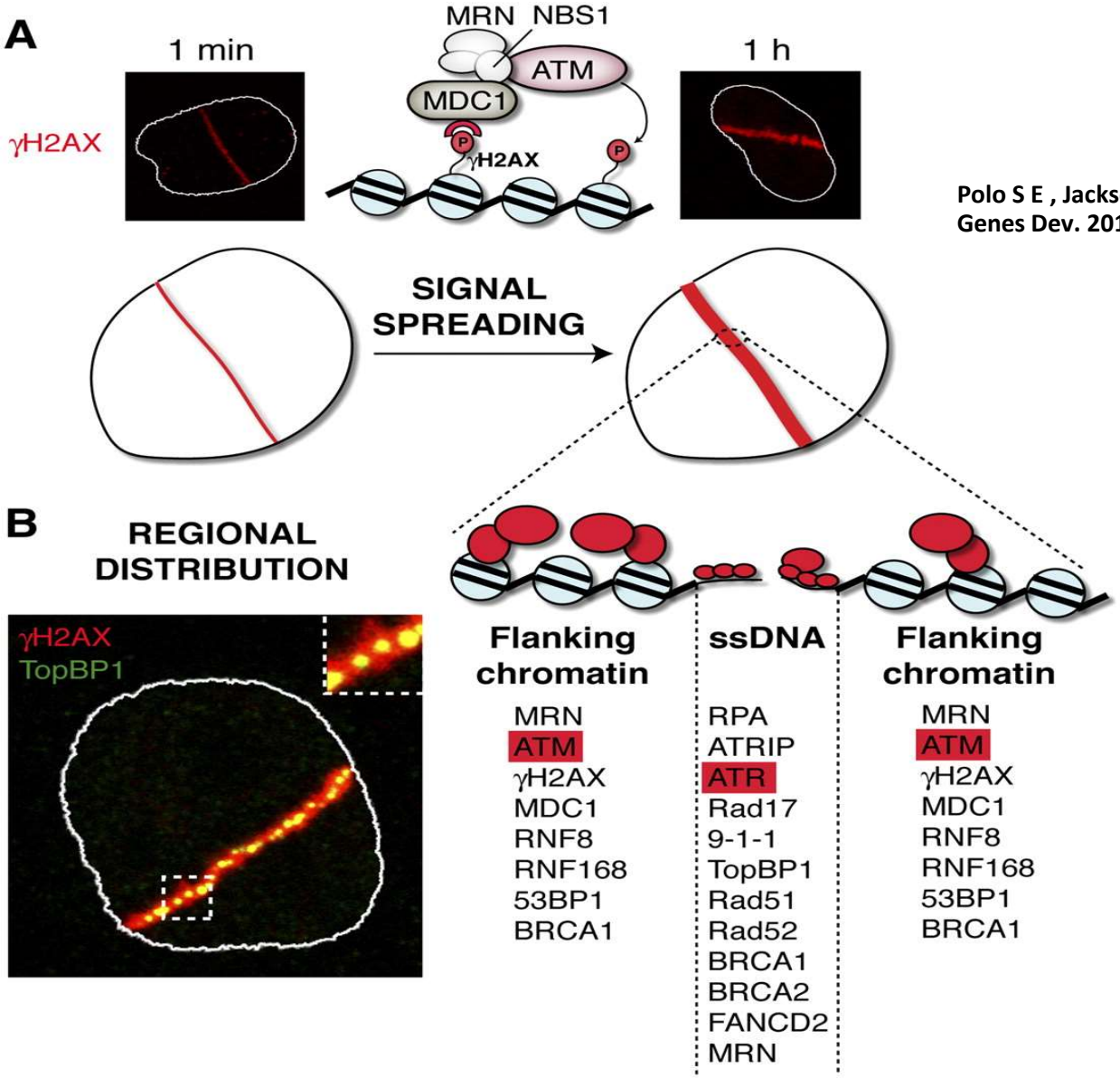
So many potential targets to visualize by immuno fluorescence

Protein dynamics to and from sites of DNA breaks.

Polo S E , Jackson S P
Genes Dev. 2011;25:409-433



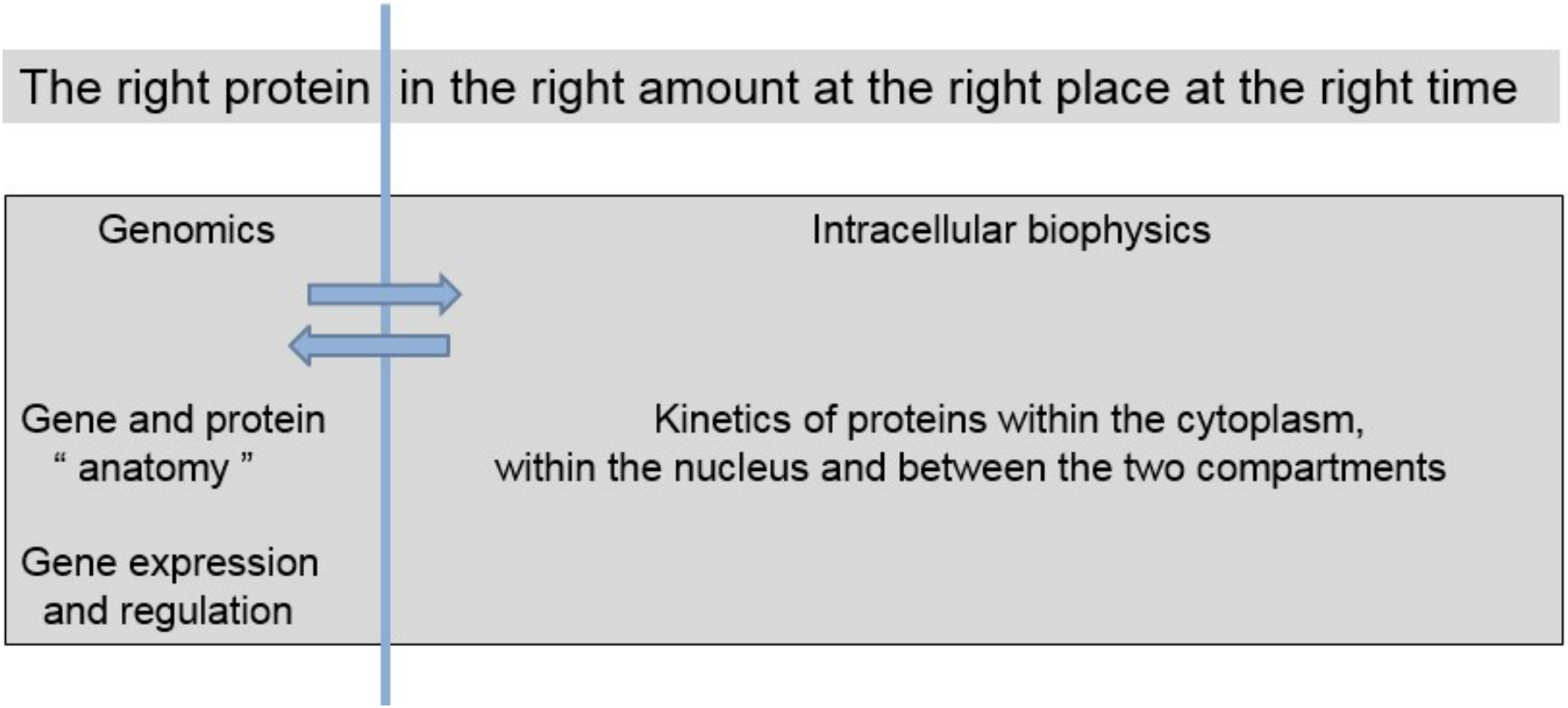
Spatial organization of DDR protein accumulation at DNA DSBs. (A) DDR signal spreading.



Polo S E , Jackson S P
Genes Dev. 2011;25:409-433

Individual response to ionizing radiation

New approach with immunofluorescence: assessment of protein kinetics inside the cells, e.g., ATM translocation from cytoplasm to nucleus



Current assays for radiosensitivity

- pATM nuclear assay (ELISA on lymphocytes for screening + extensive evaluation on skin fibroblasts)

Granzotto, Benadjaoud, Vogin et al 2016. Int J Radiat Oncol Biology Phys. 94: 450–460.
Pereira, Bodgi, Duclos et al. 2018.. Int J Radiat Oncol Biology Phys. 100:353–360.

- Radio-induced apoptosis of CD8 T lymphocytes

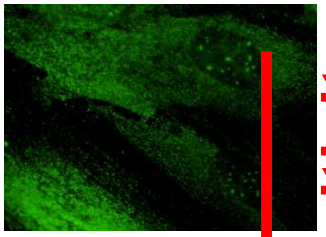
Azria, Riou, Castan et al. 2015. EBioMedicine. 2:1965–1973.

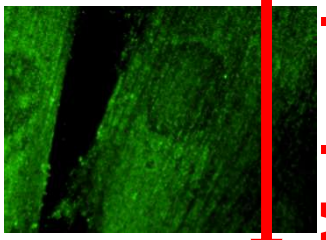
- CDKN1A / p21 gene expression

Badie et al 2008. Br J Cancer 98: 1845-51
Finnon et al. 2012. Radiother Oncol 105:329-336

Individual radiosensitivity and radiotherapy

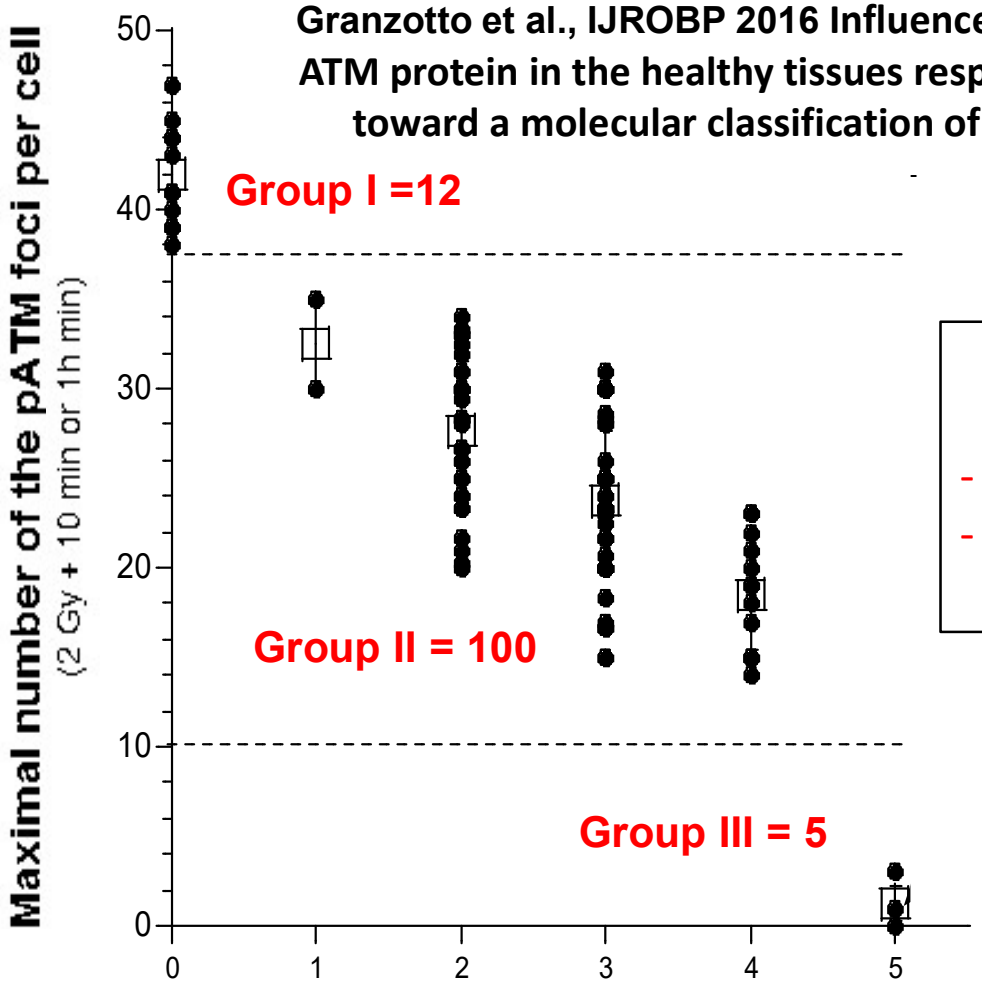
Correlation between ATM kinase activity in normal tissues and CTCAE scale severity grade : the COPERNIC Study





Molecular Radiosensitivity

Granzotto et al., IJROBP 2016 Influence of nucleo-shuttling of the ATM protein in the healthy tissues response to radiation therapy: toward a molecular classification of human radiosensitivity



A significant correlation valid :

- **for all tissues**
- **for early and late reactions**

CTCAE grade

Radiosensitivity

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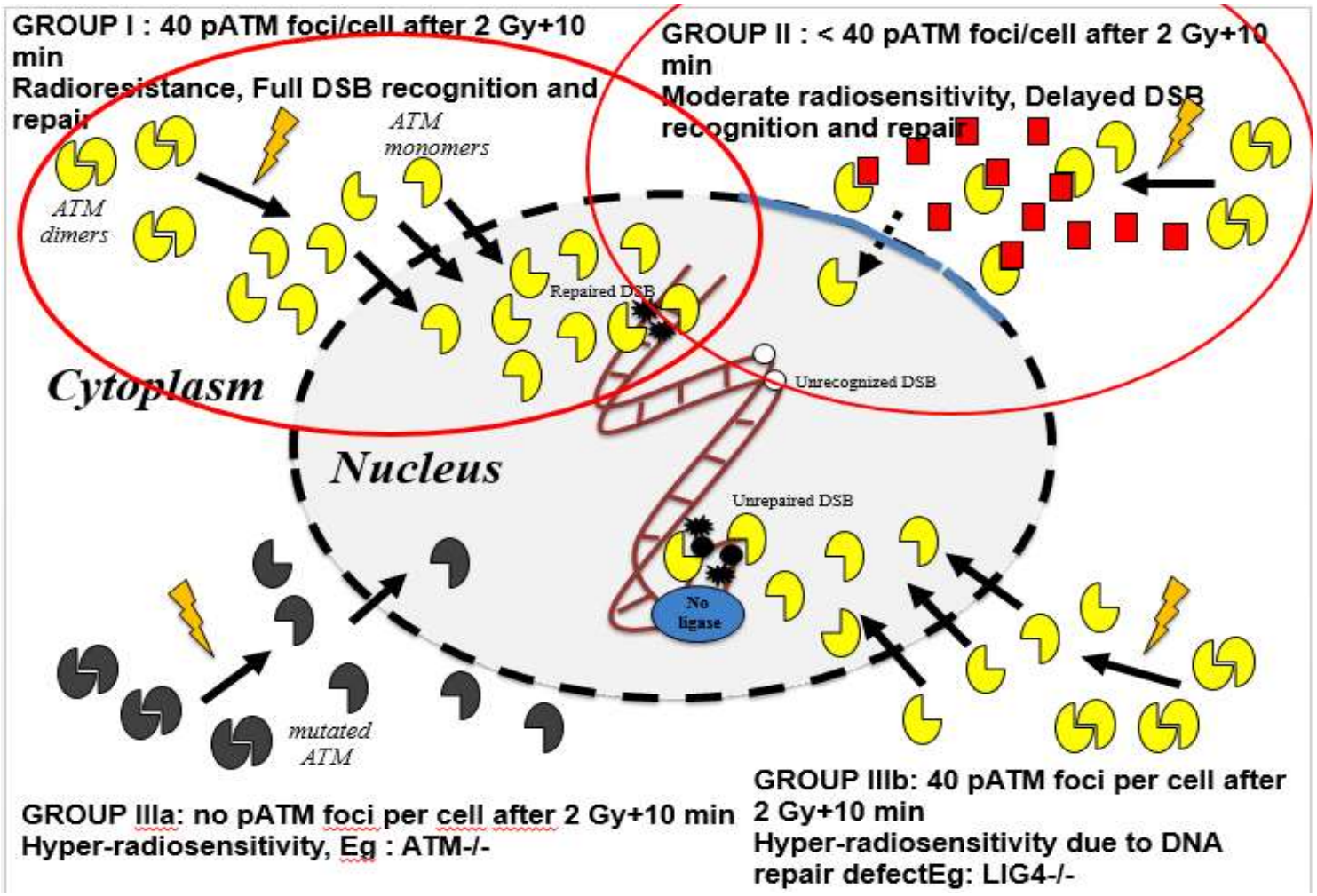
Individual response to ionizing radiation

ATM translocation (N.Foray et al – INSERM UA8)



Présenté par P.Jeggo à ERPW 2018 Rovinj Croatie pour le GT Melodi

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Individual response to ionizing radiation

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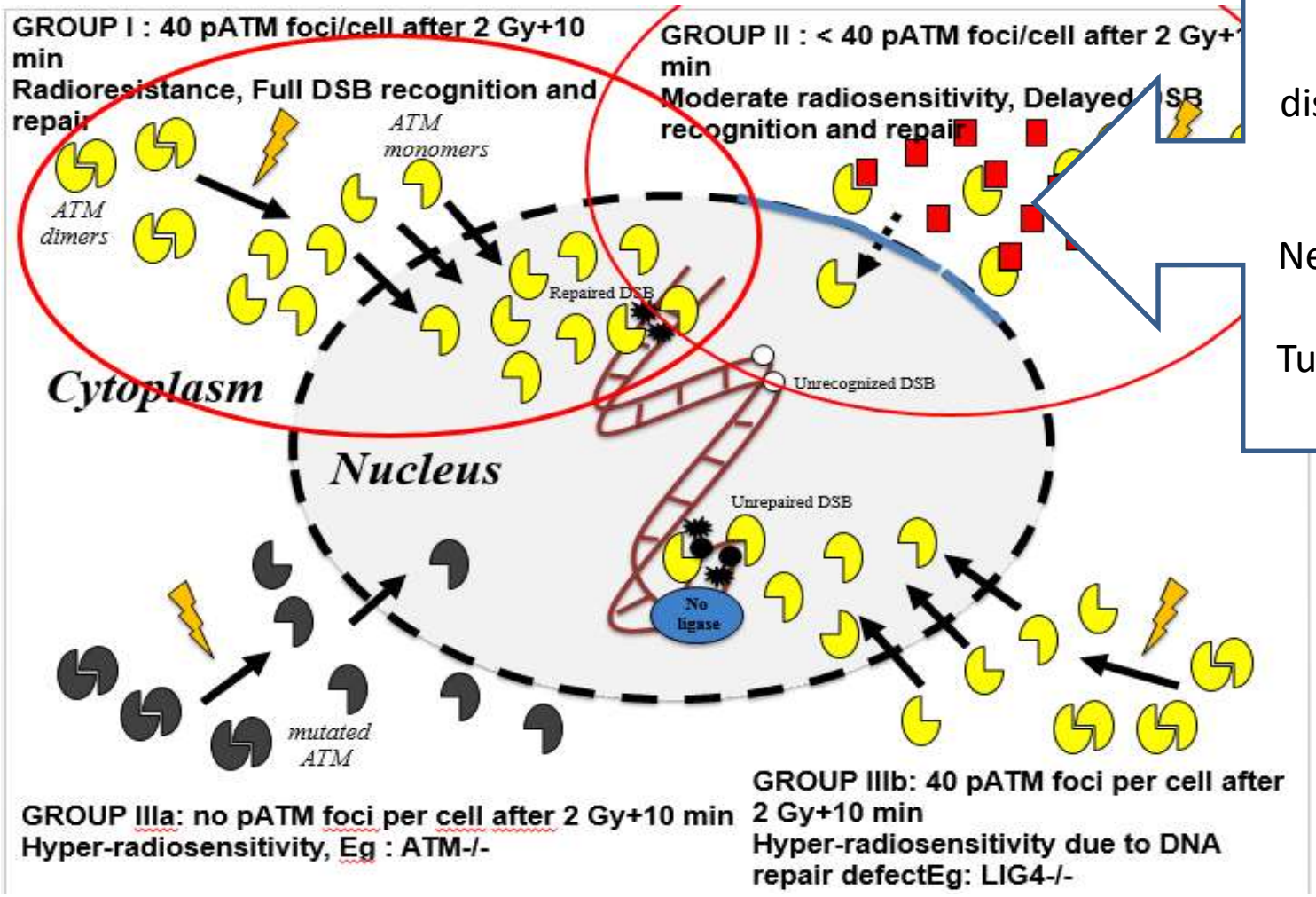
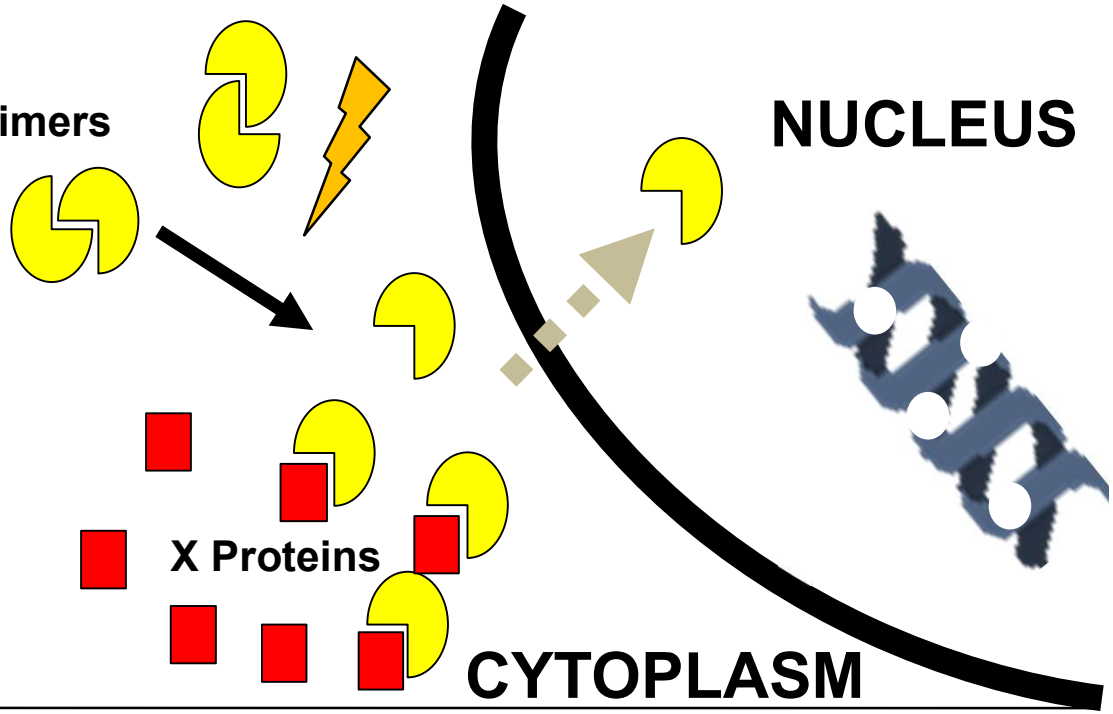


Many neuro-degenerative diseases belong to Group II : Huntington, Neurofibromatosis Xeroderma, Tuberous sclerosis, Alzheimer…

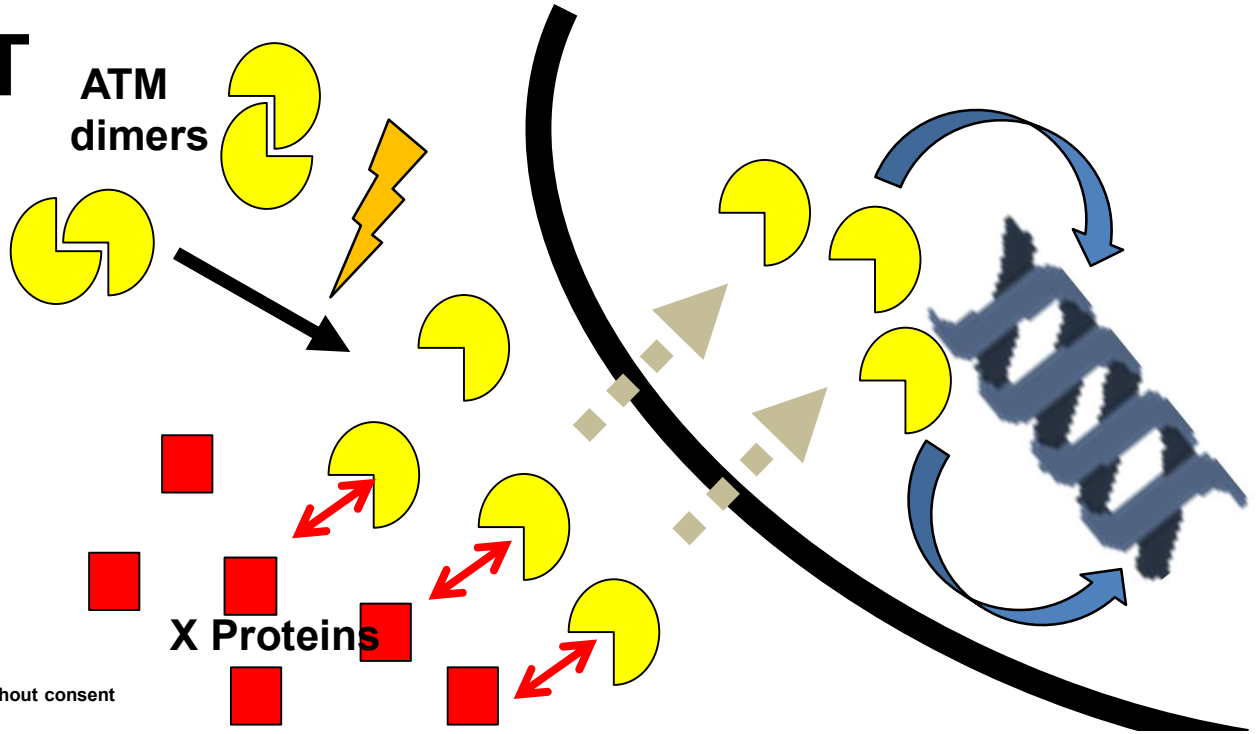
GROUP II

Delayed nucleo-shuttling of ATM
Incomplete DSB recognition
Incomplete DSB repair
Moderate radiosensitivity
High cancer risk



STATIN EFFECT

Acceleration of
nucleo-shuttling of ATM
Radioprotection

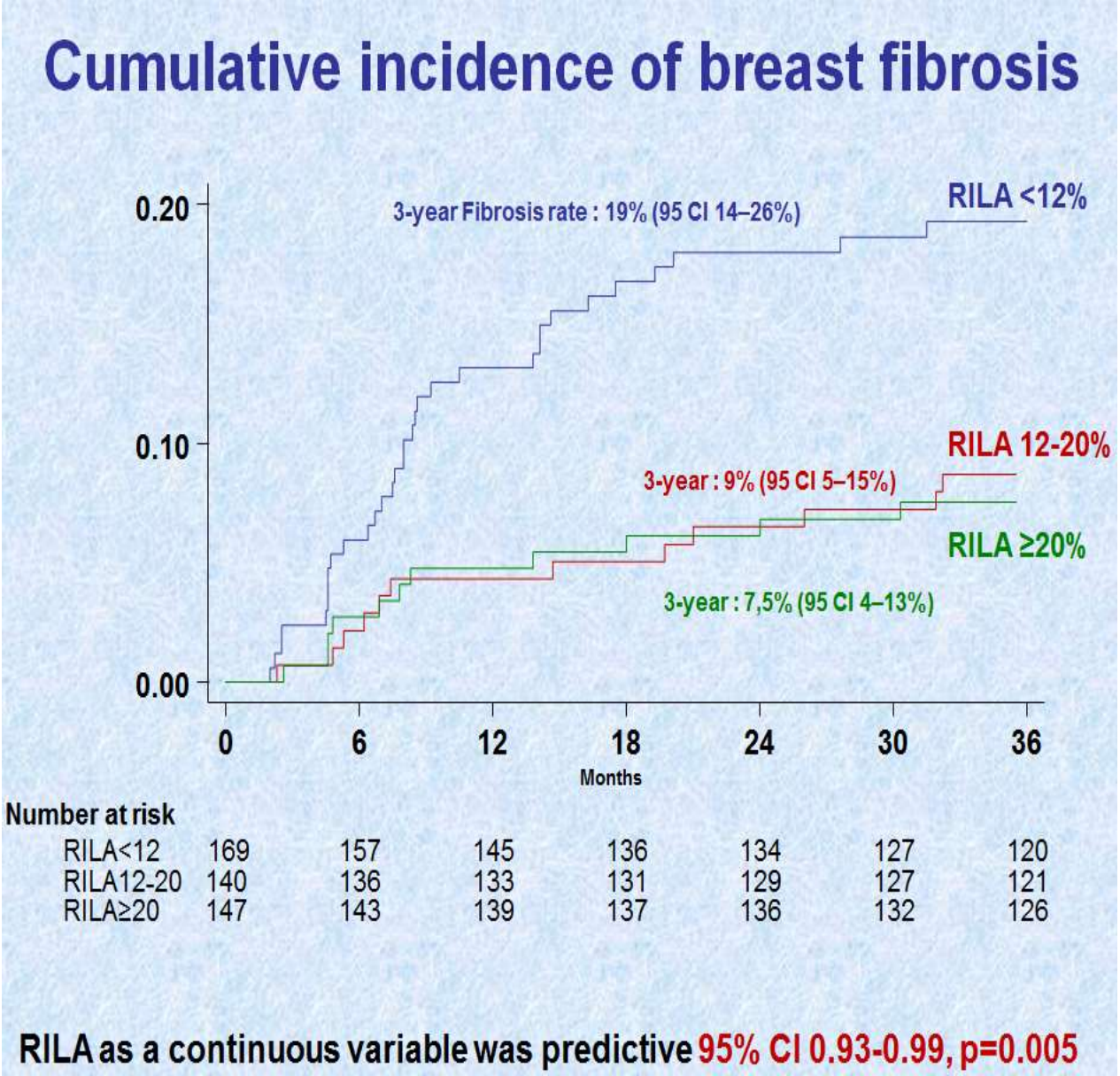


Bodgi et al. J Theor Biol 2013
Ferlazzo et al. Mol Neurobiol 2014
Granzotto et al., submitted

Radio-induced apoptosis of CD8 T-lymphocytes

RILA test

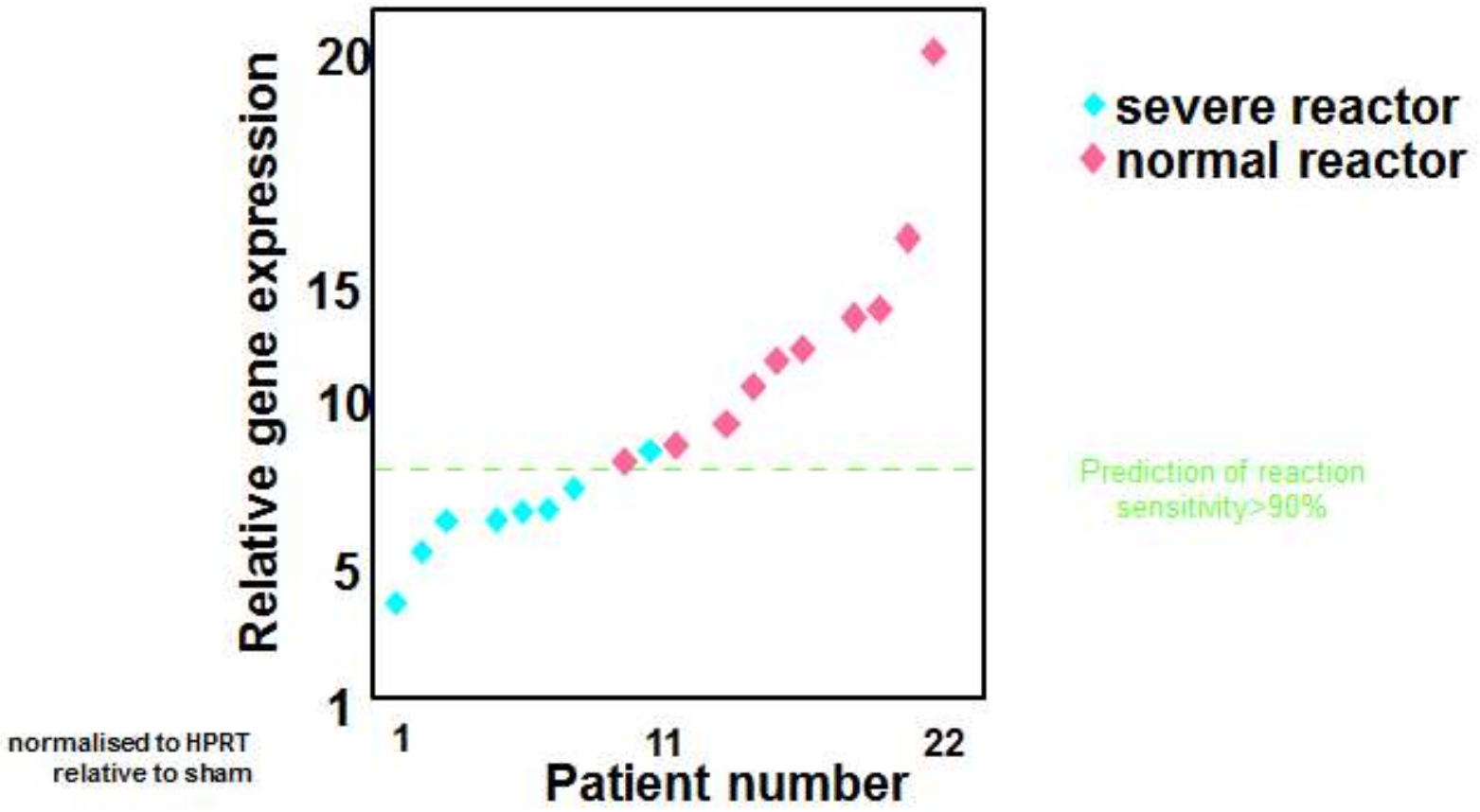
- Lymphocytes irradiated at 8 Gy
- Inverse correlation : The smaller the rate of apoptosis, the greater the radiosensitivity
- Test predictive of late complications only after radiotherapy, e.g., breast fibrosis ≥ grade 2 for a level of apoptotic lymphocytes <12 %



Azria et al, eBioMedecine 2015

Public Health England

CDKN1A as a marker of severe early radiation toxicity



Badie et al 2008, Br. J. Cancer 98: 1845-51
But also see Finnon et al 2012, Radiother Oncol. 105: 329-36

Radiosensitive patients may be radiosusceptible to cancer

- About 8% of patients treated for cancer by radiotherapy exhibit a second cancer in the field of exposure : secondary cancer or second primary cancer ?
- These patients may be radiosusceptible, i.e., cancer prone with a particular susceptibility to IR, related to the survival of altered cells

→ Another story !

Cosset JM, Hetnal M, Chargari C. Second cancers after radiotherapy: update and recommendations Radioprotection 2018, 53:101-105

Individual response at low doses IR

γH2AX foci quantified with human mammary epithelial cells exposed ex vivo in mammographic condition

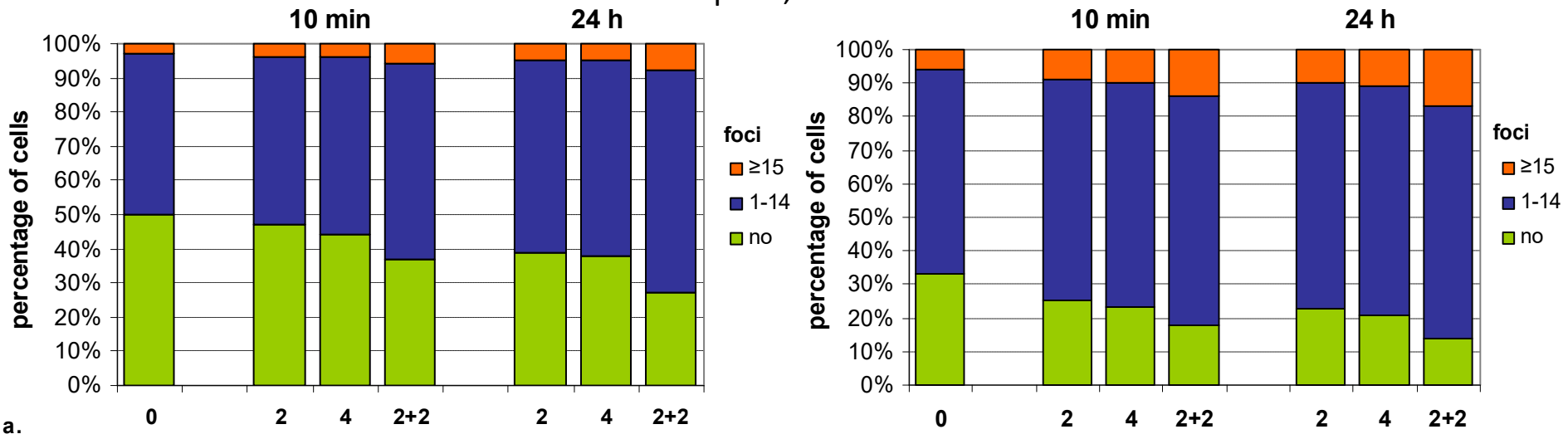
Dose effect, repetition of dose effect, effect of induction of DNA DSBs between 10 min and 24h

The 3 effects increased

Low breast cancer risk

High breast cancer family risk

p < 0,001



Exposures in mGy

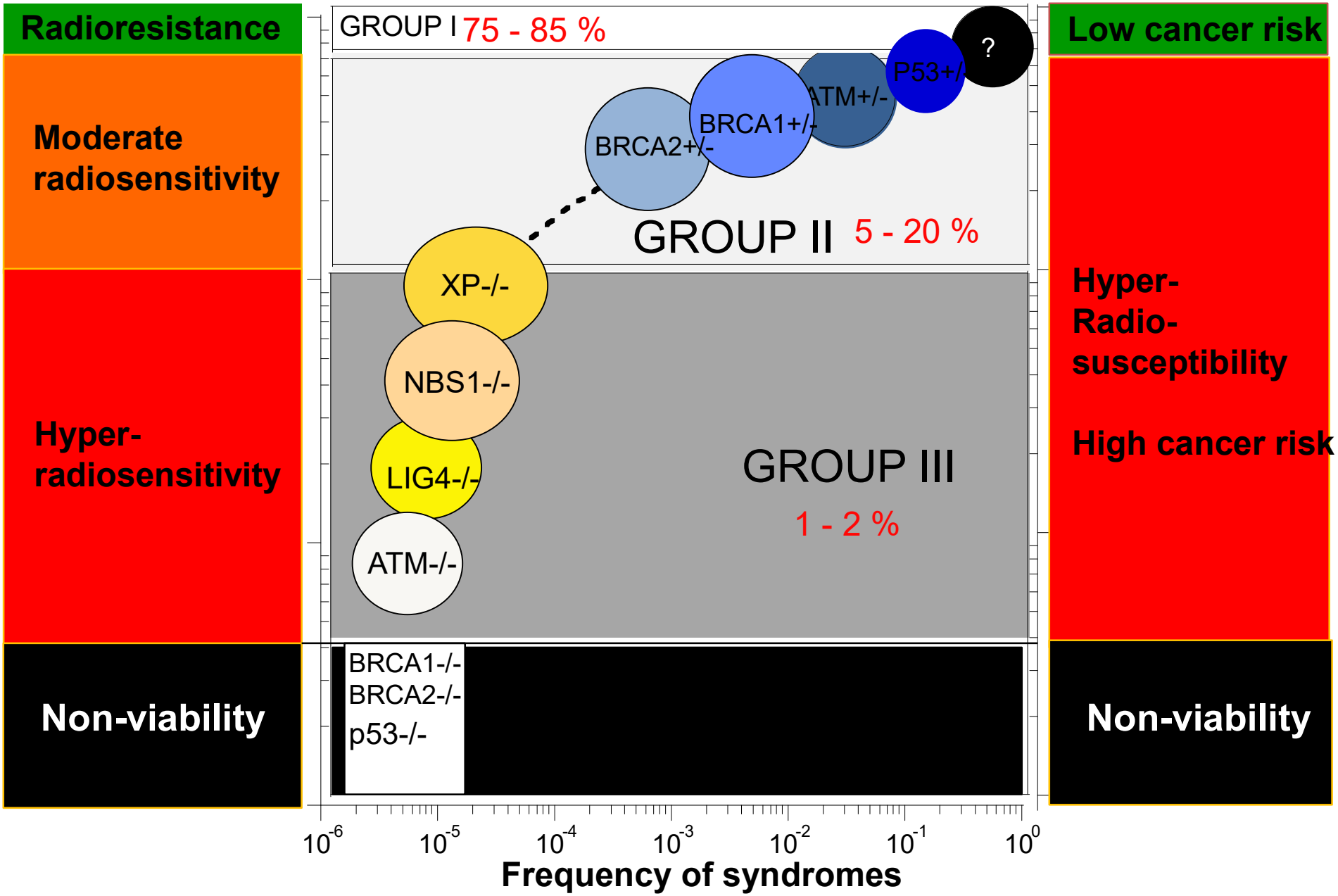
Exposures in mGy

Colin et al. 2011

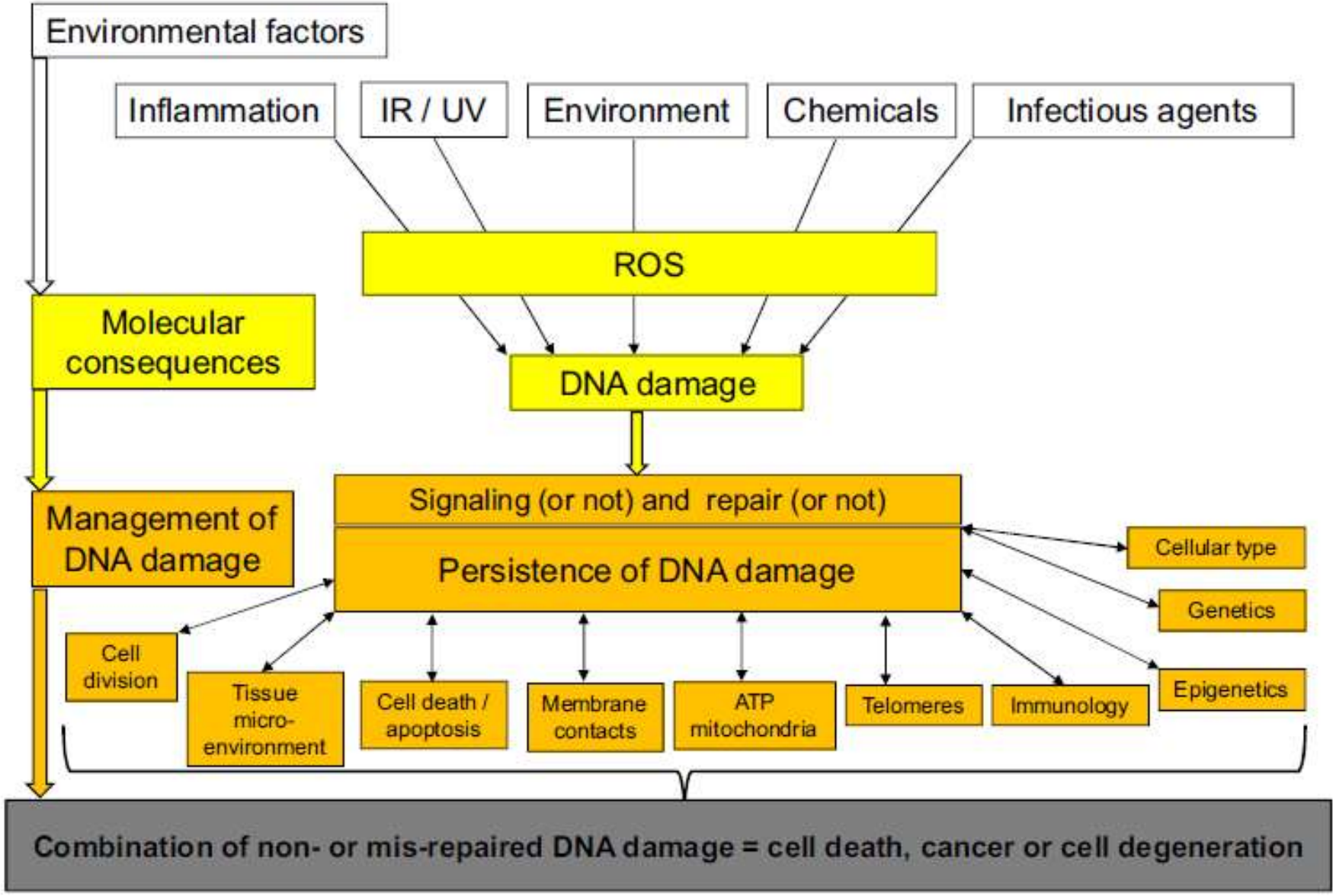
TISSUE REACTIONS

Radiosensitivity and radiosusceptibility

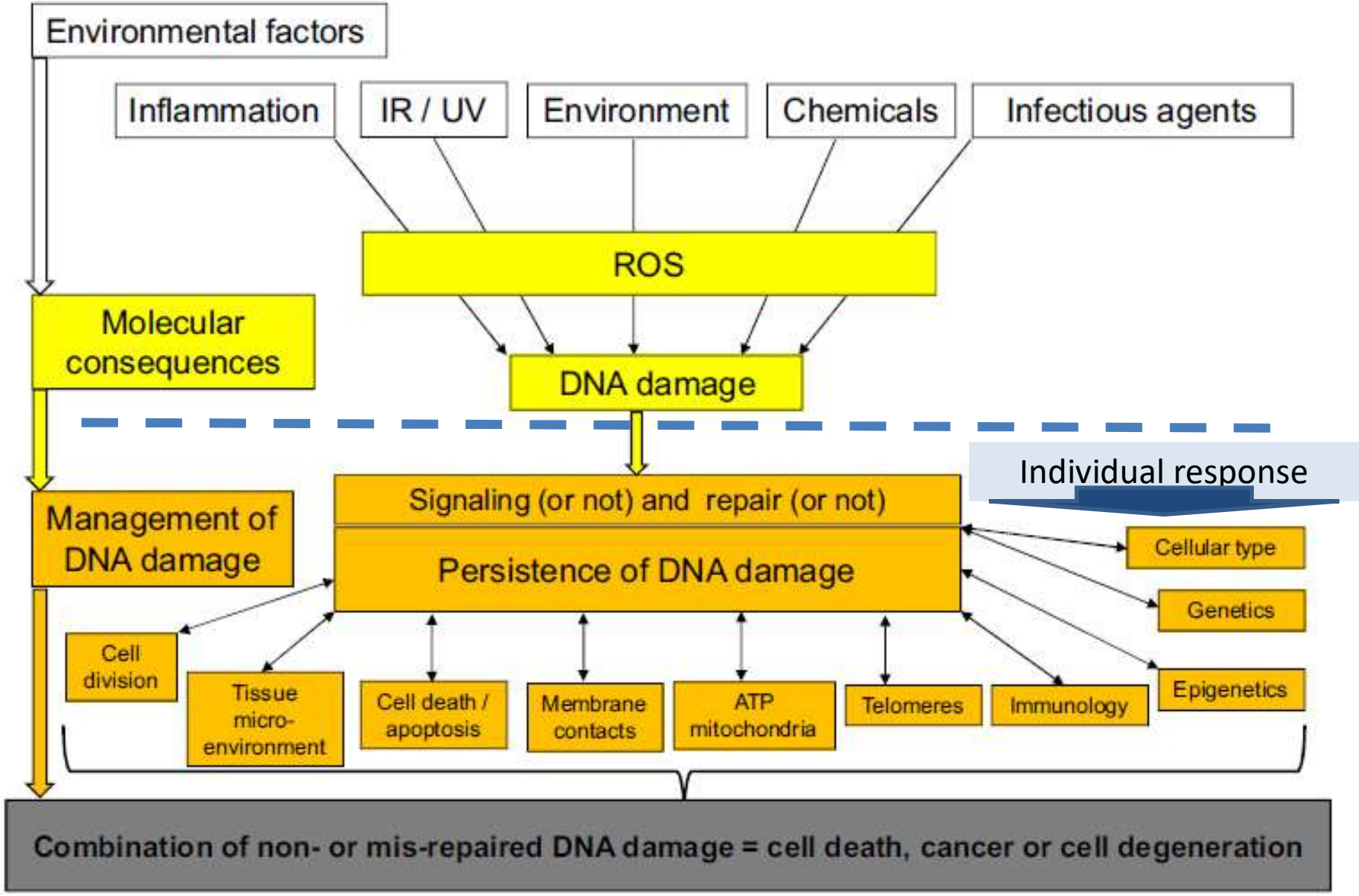
CANCER



Individual response to genotoxic agents



Individual response to genotoxic agents



Conclusion (1)

A new vision for radiological protection

Identification of patients at risk to control/decrease exposures to IR : personalized medicine and risk evaluation (ICRP TG111)

- Radiosensitive patients, to prevent or minimize the severe radio induced lesions (with alteration of quality of life) after radiotherapy, especially in children
- Radiosusceptible individuals, to anticipate cancer (prevention, screening…), notably in high family risk of cancer with no identified genes (majority of cases for breast cancer), in children, in persons exposed to radon …
- Individuals at risk of radiodegeneration to prevent the disease or slow down the evolution

Individual response to ionizing radiation

Conclusion (2)

Still a long way to go ?

In practice

- A few screening tests are already available for routine use in clinical practice
- Cross comparisons of tests are necessary on different cohorts for completing their validation
- Qualified and certified radiation biologists are necessary
- Results to be transferred to clinicians (radiation therapists…) for further decision and application

In research

- Further develop screening tests : protein kinetics / omic tools
- Design and use drugs to restore ATM translocation in neuro degenerative disease
- Understand the mechanisms of oncogenesis/degeneration (useful for cancer treatment ?)
- Enlarge the vision to all genotoxic compounds at low doses

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Thank you for your attention