UNSCEAR
United Nations Scientific Committee on the Effects of Atomic Radiation

UNSCEAR 2020/2021, Annex C BIOLOGICAL MECHANISMS RELEVANT FOR THE INFERENCE OF CANCER RISKS FROM LOW-DOSE AND LOW-DOSE-RATE RADIATION

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Aims

- To synthesize the current knowledge on biological mechanisms of radiation actions at low doses and low dose rates
- Assess their implications for understanding the processes of cancer development after exposure to ionizing radiation
- To explore the implications for dose–response relationships of radiation-induced cancers

Underpinned by a series of five specific questions formulated and agreed by the Committee to guide the development of the annex

Dose and dose-rate definitions

Low doses

those less than or equal to 100 milli-Gray (mGy) low-linear energy transfer (LET) exposure, or less than or equal to one track traversal per cell of high-LET exposure

Low dose-rates

those of 0.1 mGy/min or less low-LET exposure, or no more than one high-LET track traversal per cell per hour

Relevant prior UNSCEAR evaluations

1986 - annex B, *Dose-response relationships for radiation-induced cancer*

1993 - annexes E, *Mechanisms of radiation oncogenesis* and F, *Influence of dose and dose rate on stochastic effects of radiation*

1994 - annex B, *Adaptive responses to radiation in cells and organisms*

2000 - annexes F, *DNA repair and mutagenesis* and G, *Biological effects at low radiation doses*

2006 - annexes C, *Non-targeted and delayed effects of exposure to ionizing radiation* and D, *Effects of ionizing radiation on the immune system*

2012 – White paper, *Biological mechanisms of radiation actions at low doses*

Timeline

2016 – adopted into workplan for the Committee

Expert Group established

2017 – progress report to the Committee

2018 – first draft annex presented and discussed

2019 – second draft presented and discussed

2020 – approved annex by the Committee for publication

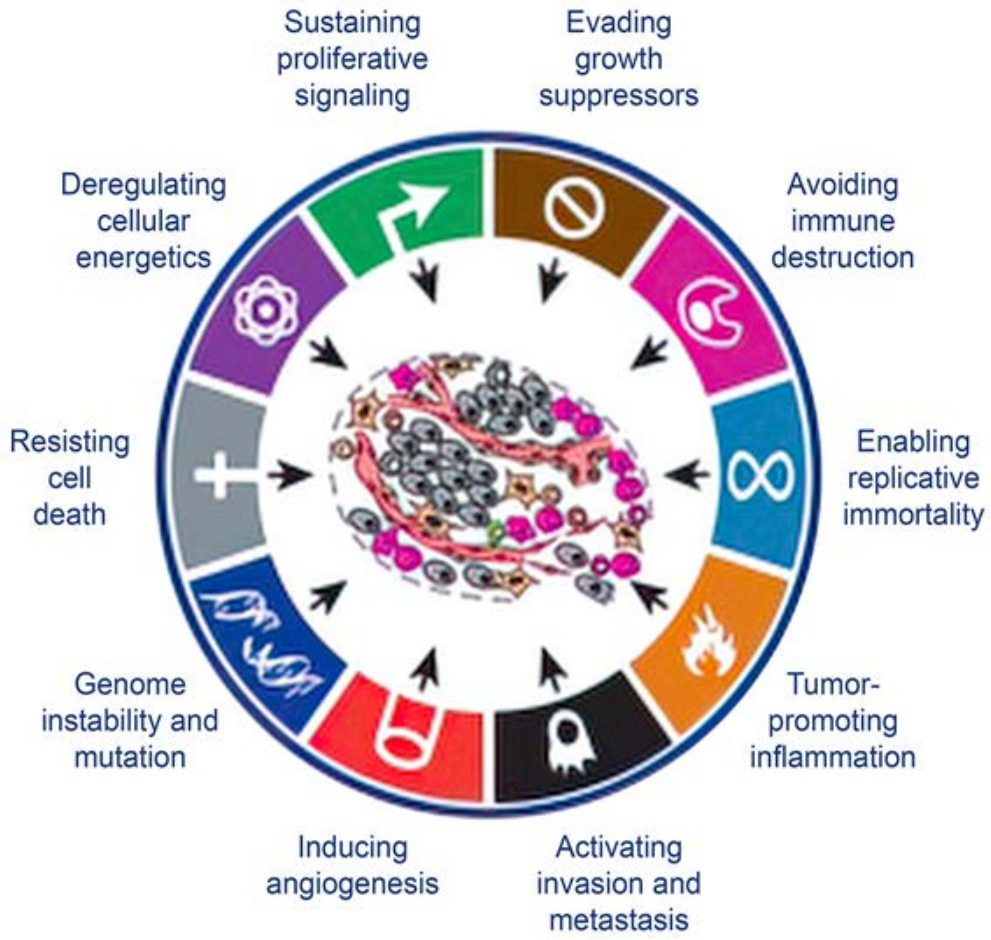
2021 – Annex published, December 2021

Expert Group

[2018 meeting]

Simon Bouffler (UK), Serge Candéias (France), Markus Eidemüller (Germany), M Prakash Hande (Singapore/India), Leon Mullenders (Netherlands/Belgium) and Gayle Woloschak (USA)

Framework for identifying relevant processes:
The Hallmarks of cancer



Adapted from Hanahan, D. and R.A. Weinberg. The hallmarks of cancer. Cell 100: 57-70 (2000).

Hanahan, D. and R.A. Weinberg. Hallmarks of cancer: The next generation. Cell 144(5): 646-674 (2011)

Areas of scientific literature considered

- DNA damage
- DNA damage signalling, chromatin remodelling and epigenetics
- Effects on other signal transduction pathways
- Gene and protein expression
- DNA repair and effects on somatic cells
- Genomic instability, bystander effects, damage/effects on non-nuclear cellular components, adaptive response and hyper-radiosensitivity
- Stem cells and target cell populations for radiation carcinogenesis
- Effects at the whole organism level

Approach to literature searches and evaluation

- PubMed used as primary source to identify relevant papers
- Main focus on papers published 2006 – 2020 (but not exclusive)
- Use of defined search terms that can be found in the annex
- Rigorous evaluation considering:
  - Is the publication original (and not a review, editorial, commentary or correspondence)?
  - Is there experimental evidence indicating that the endpoints described can be linked directly or indirectly to radiation carcinogenesis?
  - Is the experimental design adequate and free of substantial flaws, including in dosimetry?
  - Are the results statistically sound?
  - Have the results been replicated or otherwise substantiated
- Documented in an extensive Excel spreadsheet and EndNote

For which biological mechanisms and pathways is there evidence that indicates that they can affect the frequency of cancers following exposure to ionizing radiation, including at low doses and dose rates?

- very robust and reliable evidence that following the induction of DNA damage incomplete, failed or otherwise dysfunctional DNA damage responses contribute to induced mutation and chromosome damage and thereby affect the occurrence of cancers after exposures at all doses and dose-rates studied
- DNA repair activities can serve to reduce yields of mutations and rearrangements, but they are not 100% effective, do not operate in mitosis and some repair pathways may be induced or regulated by exposure
- noted that persons carrying certain variants of DNA damage response genes may be radiosensitive and at increased risk of cancer spontaneously and after radiation exposures

- There is evidence that variants of genes involved in chromatin remodelling also affect cancer risk, indicating that chromatin remodelling pathways are likely of relevance for radiation cancer risk
- Modelling studies with some support from experimental work indicate that low dose exposures can promote the growth of pre-existing pre-malignant cells and clones in tissues
- Limited evidence from occupational exposure of medical workers indicates that gene variants relating to immune functions can modulate cancer risk, whether radiation exposure per se can stimulate or suppress such cancer immunity is unclear
- There is an emerging understanding that radiation exposure, a least at moderate dose levels may stimulate tumour angiogenesis; while data at low dose levels are limited, if substantiated it may serve as a pathway to promote carcinogenesis
- Some experimental studies suggest that low doses can impact on endpoints related to tumour metastasis but the available results are mixed and inconclusive at this stage

What are the differences in utilization and/or activation of these pathways and mechanisms at low doses compared with moderate doses?

- DNA damage response operates at all dose levels, but with differences in utilization of specific pathways
- The relative importance of complex/clustered damage sites and their repair is greater at higher dose levels (and after high-LET exposures) but complex/clustered damage does occur after low-dose exposure to low-LET radiation
- Some pathways of DNA repair show inducibility such that they are up-regulated after certain exposure levels
- There are known thresholds (200-500 mGy) for the G2/M phase cell cycle checkpoint
- Some studies of gene expression following radiation exposure suggest differing responses at low as opposed to moderate/high doses but there is no consensus on the pathways specifically regulated at differing exposure levels. Also short times after irradiation, their relevance for carcinogenesis is therefore not clear
- The dose range over which the potential promotional effects of radiation on pre-malignant cells operates is unclear.
- The dose range over which cancer immunosurveillance operates is not known, but evidence arises from low dose/chronically exposed occupational groups

What evidence is available on the form of the dose–response relationships for these mechanisms?

- The dose–response relationships for mutation (LOH) and micronuclei are linear in form in the low dose range down to at least 50 and 10 mGy low-LET radiation, respectively
- The dose-response for DNA damage response activation is linear down to 10 mGy low-LET radiation
- The dose-response for the cancer immunosurveillance is not known, but the process has been observed to protect from cancer in occupationally-exposed groups, and in mice, immune system activation signalling operates at 1 mGy low-LET radiation and above
- The potential promotional effects of radiation appear to operate at 50 mGy low-LET radiation and above, but further data are required

Considering the relevant mechanisms identified, can any conclusions be drawn as to their overall influence on the dose–response relationship between cancers associated with radiation exposure at low doses compared with moderate doses?

- The knowledge of the mechanisms that affect cancer risk at low doses suggests that an overall threshold for cancer induction is unlikely, and there is evidence that the known mechanisms operate at least down to 10 mGy
- The mutational mechanism would imply a dose-risk relationship without a threshold
- At the lowest dose levels, where DNA double-strand breaks are induced in say 1 in 10 or fewer cells (around 2 mGy low-LET exposure), ROS mediated effects are likely to predominate, and these include the potential promotional action of radiation

Are there ways to link information on the biological processes and mechanisms found to be relevant to human cancer and existing epidemiological data on incidence of disease in exposed populations?

- Yes, and this review identifies two routes
- Firstly through the application of radiation-related disease biomarkers in epidemiological investigations of risk. These have the potential to reduce the time taken to obtain epidemiological data through the use of robust surrogate markers of disease when and where available and improve its accuracy by means of reducing the impact of confounding by competing causes of death and co-morbidity
- Secondly, through the integration of qualitative and quantitative biological data into mechanistic modelling of cancer risk; the modelling approaches can be used to help inform on judgements on the relevance of specific pathways for carcinogenesis

Is there evidence for tissue-specific variation in the mechanisms of response to ionizing radiation that relate to the differing sensitivity of tissues to radiogenic cancer?

- long-standing evidence that the number of mutational steps required for leukaemia is less than in the case of solid cancers
- skin stem cell populations have been found to have different responses to radiation in terms of apoptosis and this appears to relate to the higher risk of basal cell carcinoma
- stem cell populations appear to have a greater dependence on HRR than NHEJ, this may serve to provide a relative degree of protection of stem cell populations from induced mutation

Are the mechanisms that operate and can be associated with disease development similar following low- and high-LET exposures?

- Complex/clustered damage has a greater role following high-LET exposures and is more challenging to repair
- high-LET exposures readily trigger the G2/M checkpoint independent of dose
- modelling studies for lung cancer suggest that high-LET exposures may have a greater promotional effect on pre-malignant cells and clones, but this is not exclusive to high-LET exposure
- inhomogeneous distribution of radon (and progeny) in the lung leads to a protracted high-dose-rate exposure of a small population of cells, and this protracted irradiation is likely to impact on lung carcinogenesis

Directions for future research -I

- Many gaps in the evidence and knowledge on the biological mechanisms relevant for low-dose radiation cancer risk inference
- There remains a need for studies that explore the sub-100 mGy region more thoroughly and include moderate doses for the purpose of comparison at different dose levels and extrapolation between dose levels, studies using enzymatically engineered DSBs may be useful
- Better quantitative data on the induction and frequency of complex DNA damage sites, and further information on the induction of damage to mitochondria by ROS, and the specific targets within the mitochondria could be useful.
- Understand the dose/dose-rate/quality dependence of epigenetic alterations caused by radiation exposure
- Understand the dynamics of post-translational modifications (especially phosphorylations/dephosphorylations)
- Gather information on sites/genes that low-dose exposures methylate/demethylate, or otherwise epigenetically alter and the impact of these alterations on transcription
- Research is needed to improve the reproducibility and inter-comparability of results from gene/protein expression studies and to follow any changes over longer time periods

Directions for future research -II

- Provide a better understanding of the persistence of residual DNA damage and the fate of cells carrying these after milligray-level exposures
- Determine if radiation exposures do increase the occurrence of chromothripsis and the re-integration of chromosome fragments generated and the relevant exposure–response relationships
- Provide greater insight into the generation, dose-response relationship and persistence of ROS and the consequent cellular/mitochondrial effects under physiologically realistic low O_2 conditions

Directions for future research -III

- Determine whether in vivo exposures lead to persistent elevation of mutation/chromosome aberration/epi-mutation/chromothripsis frequencies that drive carcinogenesis in human or animal model systems, and to confirm if thresholds for the induction of genomic instabilities exist
- Ascertain if bystander induction of cancer occurs in humans after ionizing radiation exposure and whether bystander effects are generally cancer risk enhancing or risk reducing
- Assess the relative importance of adaptive response by comparison with the influences of other contributory risk factors

Directions for future research -IV

- Research to determine how inflammation and immune functions are affected by low-dose and low-dose-rate exposures in vivo (environmental, occupational and experimental) and how any such effects modulate cancer risk
- Further data are required from human and animal model radiation cancers, and organoid culture systems to build up a picture of the key events that convert normal cells to cancer cells, and the dose- and dose-rate–effect relationships
- Understanding the impact of low-dose and low-dose-rate exposures on later stages of carcinogenesis is also required, considering the preliminary information available on neovascularization, and endpoints related to tumour metastasis
- Mechanistic modelling studies to assess the relative importance of genomic instability/bystander effects/adaptive response, relative importance of mutation/epi-mutation, etc.
- To determine responses of relevant stem/progenitor cell populations and the role of stem cell competition

Directions for future research -V

Overall, the Committee encourages the *close multidisciplinary working* of radiobiology/epidemiology/mathematical modelling that has the potential to generate the critical data required to develop predictive models for risk inference that make use of and capitalize on all available robust and reliable knowledge of biological mechanisms and apply the knowledge to risk inference.

It will be important that *mechanisms defined using in vitro conditions are translated to in vivo conditions* in humans; both experimental and theoretical approaches can be expected to be informative

Summary of conclusions

- Little in the way of robust data could be identified that would prompt the need to change the current approach taken for low-dose radiation cancer risk inference as used for radiation protection purposes and for the purpose of comparison with other risks
- The potential contributions of phenomena such as transmissible genomic instability, bystander phenomena and adaptive response remain unclear
- There remains good justification for the use of a non-threshold model for risk inference
- However, there are ways that radiation could act that might lead to a re-evaluation of the use of a linear dose-response model to infer radiation cancer risks
- Looking to the future, the recommended approach to combine mechanistic understanding of low-dose radiation carcinogenesis with epidemiological studies is to use mathematical modelling integrating data from experimental systems

Appendix

PRINCIPLES AND CRITERIA FOR ENSURING THE QUALITY OF THE COMMITTEE'S REVIEWS OF EXPERIMENTAL STUDIES OF RADIATION EXPOSURE

- Companion to the earlier Appendix to UNSCEAR 2017, Annex A - "Principles and criteria for ensuring the quality of the Committee's reviews of epidemiological studies of radiation exposure
- Represents a strengthening of the robustness and transparency of UNSCEAR evaluations
- Provides guidance on the strengths and limitations of individual experimental studies

Thank you for your attention

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