Comments to the HLEG document:

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We have read the draft document of HLEG group with great interest. An accurate summary is given of the current knowledge and uncertainties in the context of the assessment of low-dose radiation risks. The document also proposes directions for future research to address the current ambiguities relating to the biological effects of low doses. To determine the risks of exposure to low doses of radiation emphasis is given to molecular epidemiology studies of individuals and groups of people exposed to radiation. At several places in the report (see also below) the document rightfully mentions that in addition basic and mechanistic studies will be required to fully understand the risks of radiation exposure, but these remarks are not elaborated in great detail. This is also apparent when the report is read carefully: phrases like "epidemiology" and "tissue" are used very frequently (40 and 52 times, respectively) while "repair" and "instability" are used very rarely (3 and 1 time, respectively).

- We would like to propose a few amendments and suggestions relating to the directions of research discussed in the document.

1.

We are uncertain regarding the meaning of the following sentences (p.7):
"There is, however, wide agreement that DNA damage response processes are likely to play an important role in radiation-associated cancer risk and that a variety of less well understood epigenetic factors and non-targeted effects may also be involved. Until there is a comprehensive biological understanding of carcinogenesis in general, identification and precise quantification of the particular roles of radiation remain elusive, particularly at low doses."

- This last statement could be interpreted as negative argument for any basic research in radiation biology.

2.

On p.11 the term 'systems biology' is introduced without a description of its meaning. 'Systems biology' is not well defined and as such, subject to different definitions and interpretations. Basically, two approaches are generally considered to understand the response of a cell, tissue or organism to stress including radiation:
-"Systems biology' as an approach to describe the overall set of pathways and interactions of components ('networks') within a single cell. This approach would allow understanding and predicting of the cellular response to stress such as low dose radiation.
-"Systems biology' as an approach to understand the intercellular communication between cells within a tissue.

- Our concern is that the HLEG strategy is very much focused on the tissue response rather than on single (stem) cells although both levels of 'systems understanding' are required to understand radiation induced cancer and to make meaningful predictions of risk.

3.

As mentioned in the document "Many factors have been identified that can influence the shape or the steepness of the dose-response relationship. These include differences between individuals." (p10). Individual variability in radiation sensitivity/response and its relation to genetic susceptibility might be one of the most prominent factors for low dose cancer risk. However, to assess this issue one needs to study biological effects other than cancer. We suggest that biological endpoints indicative for cancer risk (genetic damage (such as Micronuclei Chromosomal aberrations, LOH) for which quantitative assays are available (some were developed in RISC-RAD) and genomic instability (for which no quantitative measurements are available yet) should be taken into account. Particularly, data on chronic low dose exposure are lacking.

- Low dose curves for various steps in the carcinogenic process should be pursued (assays for damage transmission, epigenetic alterations and DNA damage response in surviving cells or pre neoplastic tissue (see Bartkova et al, Nature 434, 864 (2005)).
- To include experimental approaches to understand which cellular parameters influence the low dose radiaiiton response at various steps in the cancer process. Preferably one should use selected cell and tissue cancer model systems that are important for cancer/health effects in (as mentioned in the HLEG report) in a systems biology approach (see above).

4.

In general we have insufficient knowledge about the overlap between radiation sensitivity and genetically determined cancer susceptibility a question that urgently needs to be answered.

5.

As mentioned by HPA,: "opportunities have been identified where epidemiological analyses can be productively linked to laboratory studies. Given the very wide range of genetic variants that exist in the human population that might impact on low dose risk, it is highly unlikely that sufficiently powered epidemiological studies will be conducted to assess the quantitative impact of all potential risk variants. We agree that combined epidemiological and animal model studies will be of use in identifying risk variants and like to add:

- Inclusion of various <u>functional</u> assays for radiation sensitivity in these epidemiological studies will increase the power of identifying the risk factors in later genome wide association studies.
- Additional functional cohorts could come from human longevity studies (cancer susceptibility and radiation response), cancer susceptible individuals and RT patients with aberrant response.

6.

On page 15 (line 38: “Mechanistic understanding is required…..) and page 21 (line 36: More basic research will, however, be an essential component of any low dose risk….) the document indicates that in addition to epidemiology studies mechanistic research of pathways underlying the cellular responses to radiation damage is important to fully understand the risks for individuals exposed to (low) doses of radiation. Although numerous factors involved in repair and damage signaling are known, recent studies (also within the framework of Risc-Rad project) revealed novel pathways not implicated previously in radiation health risk. In addition to known mechanisms, remodeling of chromatin structure, sister-chromatid cohesion and targeting of damage to nuclear pores are crucial in maintaining genome stability. Defects in these processes are often associated with tumor formation in man.

- To improve our current understanding of factors contributing to variation in individual risk, basic research of mechanisms underlying cellular DNA damage responses will be essential component of the future research.

7.

Radiation Quality: The HELG document considers this issue to be important in order to establish quantitative judgements on their relative effectiveness for inducing cancer or other effects. What information does exist is not wholly consistent (page 15). The document also mentioned: “a critical question is how radiation quality affects the initial damage (DNA and non-DNA) and its time evolution (considering both faithful repair and mis-repair processes)’ (page 15). Moreover is has been suggested to investigate the mechanisms that governs the possible different shapes of dose effect curves and their specific dependence on radiation quality.

- Also for this issue we suggest to take into account measurements of biological endpoints indicative for cancer risk such as genetic damage (for which quantitative assays are available) and genomic instability. To our knowledge, data on low dose response relationships between biological endpoints and radiations of different quality, particularly high LET radiation (either acute or chronic exposure) are very limited.