Aging and radiation: bad companions

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Summary

Aging involves a deterioration of cell functions and changes that may predispose the cell to undergo an oncogenic transformation. The carcinogenic risks following radiation exposure rise with age among adults. Increasing inflammatory response, loss of oxidant/antioxidant equilibrium, ongoing telomere attrition, decline in the DNA damage response efficiency, and deleterious nuclear organization are age-related cellular changes that trigger a serious threat to genomic integrity. In this review, we discuss the mechanistic interplay between all these factors, providing an integrated view of how they contribute to the observed age-related increase in radiation sensitivity. As life expectancy increases and so does the medical intervention, it is important to highlight the benefits of radiation protection in the elderly. Thus, a deep understanding of the mechanistic processes confining the threat of aging-related radiosensitivity is currently of foremost relevance.

Key words: chromatin organization; DNA repair; nuclear envelope; oxidative stress; replicative senescence; radiosensitivity.

Introduction

Age at the time of radiation exposure is one of the main factors involved in radiation-induced cancer. Individuals exposed at early ages are the most radiosensitive as the primary damage has a longer latent phase to outbreak into cancer. After that, sensitivity to radiation decreases until maturity, but it increases again at older ages. Epidemiological evidence for such age-dependent radiosensitivity variation has been reported in different studies. In the Life Span Study cohort of the Japanese atomic bomb survivors, the excess relative risks (ERRs) for radiation-induced cancers as a function of age at exposure were examined (Shuryak et al., 2010). As expected, the ERR for cancer induction was higher during childhood and decreased progressively at exposure ages of 30–40. Surprisingly, the ERR of developing solid tumors ramped up again for exposure ages higher than 40 years old (Shuryak et al., 2010). Richardson and Wing found a similar tendency among the Oak Ridge Y-12 uranium processing plant workers (Richardson & Wing, 1999). They observed that the radiation doses received after the age of 45 showed a stronger association with cancer mortality than those received at younger ages. All these findings suggest that the radiation sensitivity, measured in terms of carcinogenic events, increases with age among adults after age of 40–45. It has been suggested that this bimodal distribution reflects that radiation risks after exposure at early ages are related to initiation of malignant processes, whereas radiation risks after exposure at later ages are mostly associated with the promotion of pre-existing premalignant cells (Shuryak et al., 2010). Not only radiosensitivity, but also the percentage of individuals undergoing medical imaging procedures becomes significantly greater with age, and so does the cumulative effective dose from these procedures (Fazel et al., 2009). Finally, life expectancy has remarkably increased in the last decades and thus, radiation-induced tumors at older ages have more time to develop and progress.

Therefore, understanding the age-related changes that may compromise the population after radiation exposure becomes increasingly relevant. In this review, we will focus on four cellular processes: oxidative stress, telomere attrition, DNA repair, and inflammatory response, and we will provide an integrated view of how these four mechanisms may contribute to the relationship between aging and radiosensitivity.

Oxidative stress arising from aging and radiation

Lessons from the free radical aging theory

Reactive oxygen species (ROS) can be endogenously generated by the normal cellular metabolism or exogenously originated by exposure to radiation and chemical compounds (Valko et al., 2007). ROS may form compounds, such as hydroxyl radicals or hydrogen peroxide, which could initiate harmful chemical reactions in cells. High levels of ROS can cause damage to macromolecules, such as lipids, nucleic acids, and proteins. Lipid peroxidation is one of the early steps of this damaging process, followed by oxidation of nitrogenated bases. Damage to the DNA, mainly strand breaks and DNA cross-linking, is a source of mutations in the cell genome. In addition, practically, every amino acid in a protein can be oxidized by ROS. The harmful effects of ROS are termed as oxidative stress. However, the damage that ROS can induce to the cell not only depends on their concentration but also on the equilibrium between ROS and the antioxidant species. Oxidative stress is generated when there is a loss of pro-oxidant–antioxidant equilibrium, thus altering and damaging many intracellular molecules, including DNA, RNA, lipids, and proteins (Veselkova et al., 2012). Oxidative stress is important from a biomedical perspective because it is linked to a wide variety of human diseases, such as neurodegenerative and inflammatory diseases, as well as cancer. ROS can promote many aspects of tumor development and progression at different levels: cellular proliferation, evasion of apoptosis, tissue metastasis and invasion, as well as angiogenesis (reviewed in Sosa et al., 2013).
As ROS constitute a persistent source of DNA damage, they were assumed to contribute to the age-related deterioration of functions in the organism. The oldest theory that relates aging with ROS was proposed more than 50 years ago and postulates that the accumulation of ROS and the subsequent oxidative damage along the lifespan of aerobic organisms triggers the aging process (Harman, 1956, 1992). Recently, this theory has been severely criticized by some authors, who claim that the correlation between oxidative damage and the aged phenotype, even if it exists, does not imply causation (Buffenstein et al., 2008; Pérez et al., 2009; Lapointe & Hekimi, 2010). While entering in this debate is beyond the scope of this article, some points of the theory are relevant to this discussion. First, contributing to aging or not, ROS accumulation was found to be higher in aged cells in comparison to their young counterparts (Ku et al., 1993). Secondly, in addition to the increased ROS, impaired antioxidative enzymatic activities with aging have also been described. Antioxidant enzymes such as superoxide dismutase, catalase and glutathione peroxidase showed a decline in their activities and/or gene expression with age (Andersen et al., 1997; Inal et al., 2001). Nowadays, the free radical aging theory seems to evolve toward a more conservative position, which states that there is an increasing imbalance between the number of oxidant and antioxidant species with age (Junqueira et al., 2004). This imbalance may lead to age-dependent oxidative stress that compromises both cellular structures and homeostasis (Kregel & Zhang, 2007; Liu & Xu, 2011).

Ionizing radiation worsens the already unbalanced oxidant–antioxidant status in aging cells

Ionizing radiation can have various effects at the cellular level. When radiation is absorbed by the cell, damage can occur either by direct or indirect action. In a direct action, a secondary electron, which results from photon absorption, interacts with and is absorbed by a biomolecule, such as DNA, RNA, or proteins. In an indirect action, ionizing radiation impairs or damages cells by creating free radicals (Riley, 1994). Radiation results in the high local production of ROS attributable to chemical interactions between high-energy electrons, photons, and the molecular targets of oxygen and water within cells. Radiation can also produce ROS through signaling processes that evolve in their release from mitochondria. It has been proposed that intrinsic sensitivity of cells to radiation is dependent on the subsequent ROS generation as they can induce mutations in the DNA (Seong et al., 2010). Irradiating aged cells that already contain an increased amount of ROS would unequivocally contribute to overload the antioxidant systems responsible for eliminating the excess of oxygen metabolites (Fig. 1). Importantly, in an attempt to estimate the effect of aging on radiation-dependent ROS generation, Kasapovic et al. (2009) provided evidence that older women had a decreased antioxidant capacity and subsequent higher oxidative damage. The activity of the different antioxidant enzymes and concentration of lipid hydroperoxides were measured in blood cells of two groups of breast cancer patients with different ages, to estimate their capacity to eliminate H2O2 formed after radiotherapy. A decreased antioxidant capacity and an increased lipid peroxidation were observed in the older patient group. The authors concluded that radiotherapy promotes further oxidative shift, which in turn potentiates the already existing chronic oxidative stress linked to breast cancer and aging, resulting in a further increase of mutagenic potential. Similarly, antioxidant protection against curative and palliative doses of ionizing radiation in human blood was reported to decrease with aging (Kasapovic et al., 2012). Thus, radiation may cause a major impact in the elderly, whose aging cells already naturally showed an impaired redox system. In line with this, in vivo experimental studies on animal models have shown that adding antioxidant supplements to the diet can minimize the effects of ionizing radiation later on (Weiss & Landauer, 2003). Vice versa, approaches were achieved with knocked-down cytoglobin (a vertebrate globin that scavenges ROS), which made glioma cells more sensitive to radiation (Fang et al., 2011). Cytoglobin overexpression resulted in protection of human neuroblastoma cells against oxidative stress-induced cell death (Fordel et al., 2006). These studies exemplify a link between radiosensitivity and oxidative stress regulation systems that can affect the radiosensitivity of aging cells.

In summary, irradiating aging cells that already display an unbalanced oxidant–antioxidant status would unequivocally contribute to overload the antioxidant systems (Fig. 1). In aging cells, when ROS production exceeds the antioxidant defense capacity of the cell, excess of oxidative stress occurs and induces damage to the DNA, proteins, and membrane lipids. In summary, during aging, radiation further contributes to ROS generation, increasing the possibilities of oncogenic transformation.

Telomeres: effects of the age-related attrition in radiosensitivity

Telomere attrition with age

Telomeres are DNA-protein structures at the end of the chromosomes, protecting them from exonucleolytic degradation, homologous recombination, and nonhomologous end joining. In the absence of balancing mechanisms, telomeric DNA shortens with each DNA replication. Although telomerase can add new telomeric DNA to the ends of the chromosomes, its activity is basically restricted to ovaries, testes, and highly proliferative tissues such as activated lymphocytes and adult tissue stem cells (Colgin & Reddel, 1999). Therefore, most somatic cells in human tissues have insufficient levels of telomerase activity, and telomere attrition occurs in parallel with aging in rapidly proliferating cells of the skin, gastrointestinal system, and blood. Some studies have established that the mean leukocyte telomere length is inversely associated with age and it declines between 20 and 40 bp per year (Brouillette et al., 2003, 2007; Fitzpatrick et al., 2007). Once a critical shortened telomere length is attained, it triggers a state of permanent growth arrest called replicative senescence (Harley et al., 1992). Replicative senescence is induced by activating a DNA damage response similar to that caused by DNA double-strand breaks (d’Adda di Fagagna et al., 2003) and has been postulated as a tumor suppressor mechanism. In fact, malignant progression occurs when cells acquire mutations, primarily in the p53 or Rb pathways, allowing them to overcome this telomere-dependent proliferative arrest. When aging cells bearing critically short telomeres ignore the senescence-inducing signals, or escape from the senescence-growth arrest, their telomeres keep getting shorter and dangerously dysfunctional in terms of genomic instability.

Telomere attrition and genomic instability

Cell proliferation beyond replicative senescence leads to uncapped chromosomes that can fuse with each other or with its sister chromatid after DNA replication (Soler et al., 2005). Such unstable chromosome configurations can set up fusion–bridge–breakage cycles, which are prone to produce rapid and important changes in gene dosage, thus linking telomere dysfunction and chromosome instability (Fig. 2). Carcinogenesis is particularly induced when the cellular response to telomere attrition is reduced due to cell cycle checkpoint defects, as demonstrated by studies carried out in mice with impaired p53 function.
A dramatic increase in the incidence of tumors, specifically carcinomas, has been observed in hTERT/C0/C0p53+/C0 mice with short telomeres. Indeed, the cytogenetic characteristics of these tumors reproduced those identified in human carcinomas, showing a high frequency of unbalanced chromosome rearrangements (Artandi et al., 2000). Of note, mutations and deletions of the TP53 tumor suppressor gene encoding p53 are very frequent in human carcinomas (Negrini et al., 2010) and remarkably associated with organismal aging and cancer incidence (Richardson, 2013). In fact, several studies have reported an increase of DNA damage, mutations, and genome instability related to age (Burhans & Weinberger, 2007; Lushnikova et al., 2011; De Magalhães, 2013; López-Otín et al., 2013). Additionally, there might exist an age-related deficiency of the senescence response efficacy mechanisms that could lead to the proliferation of cells with critically short telomeres (Feng et al., 2007). And finally, the most direct evidence that telomere dysfunction is an important component of the genomic instability observed in human cancer comes from studies using a PCR-based assay to detect and analyze telomere fusions. Using a telomere-associated repeat fusion PCR, Tanaka et al. (2012) reported that human breast premalignant and malignant lesions, but not normal breast tissues, contained telomere fusions. Altogether, these studies support the notion that age-dependent telomere attrition in a cell environment with impaired cell cycle checkpoints contributes to human carcinogenesis in the elderly (Meeker & Hicks, 2004; Negrini et al., 2010).

Telomere attrition raises radiosensitivity

Telomeric dysfunction not only underlies the initiation of carcinomas but also relates to radiosensitivity. Cells with short telomeres are more radiosensitive than their long telomere counterparts (Goytisolo et al., 2000; Wong et al., 2000). Studies in irradiated embryonic fibroblasts from telomerase-deficient Terc−/− mice provided an important clue to understand the basis of the increased sensitivity of this mouse model. Latre et al. (2003) observed that chromosomes with unprotected ends fuse, not only to one another, but also to radio-induced DNA double-strand breaks (DSBs) (Latre et al., 2003). Thus, shortened telomeres provide radiation-induced DSBs with a new joining possibility. If aging cells fail to trigger replicative senescence (Feng et al., 2007), their uncapped chromosomes will offer additional rejoining opportunities that may increase improper repair of radiation-induced breaks (Fig. 2). Thus, if aging alone can induce chromosome instability when eroded telomeres meet checkpoint impairments, radiation would exacerbate this scenario by further compromising genomic stability in older organisms.

Impaired DNA repair of the aging cells

DNA is constantly under the attack of both endogenous and environmental agents. Given the importance of its preservation, cells have developed several mechanisms, commonly known as DNA damage response (DDR), for fighting against these threats. These mechanisms detect the DNA damage and mediate its repair while arresting the cell cycle to avoid DNA replication or segregation (Jackson & Bartek, 2010). When DNA damage persists unrepaired, DDR can trigger cell death by apoptosis or halt cell proliferation through induction of senescence. Among all different types of DNA lesions, DSBs are highly deleterious and their presence activates the DDR surveillance system (Bekker-Jensen & Mailand, 2010). DSBs can arise from ionizing radiation, oxidative stress, or replication stress, but can also be formed during genetically programmed processes such as meiotic recombination in germ cells and V(D)J recombination in developing lymphocytes (Wyman & Kanaar, 2006). The two main pathways responsible for DSB repair are...
nonhomologous end joining (NHEJ) (Lieber, 2008) and homologous recombination (HR) (San Filippo et al., 2008). Before repair is happening, the DSB is signaled by the DDR, resulting in the recruitment of proteins involved in any repair pathways. Phosphorylation of the histone variant H2AX (γH2AX) by ATM and DNA-PKcs represents the first event of the signaling cascade (Rogakou et al., 1998). γH2AX enables the binding of additional DDR factors, such as MDC1, BRCA1, and 53BP1. H2AX phosphorylation takes place at the DSB site shortly after its formation and disappears at its resolution. The phosphorylated histone H2AX is microscopically visible by immunostaining, which makes γH2AX foci a widely used surrogate to measure DSBs. We focus this part of the review on the impact of aging in proper DSB resolution (Fig. 3).

DNA damage response efficiency declines with age

It has been reported that aging cells have increased amounts of γH2AX foci as compared with younger cells, suggesting that they accumulate a higher amount of unresolved DSBs, seen as increasing γH2AX levels with age (Sedelnikova et al., 2008; Joyce et al., 2011; Rübe et al., 2011). Not only spontaneous foci are more abundant in these cells, but also a significant induction of damage has been observed following exposure to low doses of radiation in aging cells. In fact, we reported that X-ray doses equivalent to those applied to the breast surface after a single mammogram exploration, resulted in more γH2AX foci in the in vitro aged human mammary epithelial cells than in their young counterparts (Hernández et al., 2013). In our work, cells were transduced with hTERT, discarding that telomere attrition and its subsequent increase of misrejoining events could account for the increase in γH2AX foci. In addition, in vitro aged cells also showed a delay in the recruitment of the DDR protein 53BP1 to the damage site (Hernández et al., 2013). In line with these results, it had been previously proposed that recruitment rates of DNA repair proteins at DSB sites after irradiation might be inversely correlated with donor age (Sedelnikova et al., 2008). It has been reported that age may diminish the effectiveness of the two main DSB repair pathways (NHEJ and HR). Using plasmid ligation methodologies, a recent study has shown that B lymphocytes from old mice show poor NHEJ repair efficiency and increased misrepair compared with younger mice (Puthiyaveetil & Caudell, 2013). Using a similar approach, Seluanov et al. (2004) demonstrated that NHEJ efficiency is reduced in aging cells compared with young cells. The same authors also observed that the levels of Ku—a key component of the NHEJ pathway responsible for keeping close together the two broken DNA ends—were diminished in senescent cells (Seluanov et al., 2007). Reduced efficiency of NHEJ in the elderly might result in persistent DSBs leading to misrejoining events and genomic instability, and also in impaired V(D)J recombination, which could contribute to reduce the immune cell repository and thus the immune system (Puthiyaveetil & Caudell, 2013). As the immune system uses its diverse antigen receptor repertoire to prevent tumor formation and progression, NHEJ impairment could eventually lead to a carcinogenic prone scenario (Li et al., 2011).

Fig. 2 Telomere attrition in aging cells. Ionizing radiation induces new DNA double-strand breaks (DSBs), and thus, new opportunities for the uncapped chromosomes undergo unfaithful repair. When aging cells skip replicative senescence, they display a greater number of uncapped chromosomes, which are prone to produce rearrangements. They can undergo end-to-end fusions and DSB-end fusions between different chromosomes. When the two centromeres are pulled in different directions, dicentric chromosome can break, and this breakage results in further fusions followed by other bridges and, again, new breaks will arise. This process is known as the breakage-fusion-bridge cycle (BFB cycles), which leads to broad DNA amplification and progressive terminal deletions. Any of these outcomes lead to a rise in chromosome instability, which, in turn, can initiate or promote a carcinogenic process.
Similarly to NHEJ, recent reports also suggest that the HR pathway is impaired with aging. Mao et al. showed that presenescent cells displayed severe difficulties in recruiting RAD51 to DNA damage sites after irradiation (Mao et al., 2012; Chowdhury et al., 2013). RAD51 is responsible for mediating the invasion of the sister chromatid to find homologous sequence partners, thus representing a key component of the HR pathway. Intriguingly, exogenous incorporation of RAD51 rescued the repair ability in middle-aged cells, but failed to do so in presenescent cells (Mao et al., 2012), suggesting that in the oldest cells there must be other factors responsible for the DNA repair defects. To sum up, when analyzing age-related radiosensitivity, DSB repair pathways impairment must be considered. Although the exact nature of age-related misrepair remains unknown, emerging evidence points at DNA repair proteins recruitment to the damaged DNA at the nucleus as promising targets for this yet unexplored research area.

DNA damage, inflammation, and aging

Cells respond to foreign DNA introduced in the cytoplasm by triggering innate immune responses, which are not specific to a particular pathogen in the way adaptive immune responses are. The accumulation of bacterial or viral double-stranded DNA in endosomes and DNA by-products derived from retroviruses in the cytosol triggers immune activation (Hemmi et al., 2000; Ishii et al., 2006; Stetson & Medzhitov, 2006). Of relevance, as DNA breaks can be generated during viral integration, the nucleus is not invisible to immune DNA sensors. Consequently, nuclear DNA damage triggers a chronic autoinflammatory response (Karakasilioti et al., 2013). In this sense, etoposide-induced DSBs in the nuclear DNA stimulate a cascade of proinflammatory signals through the induction and action of interferon cytokines (Brzostek-Racine et al., 2011). The mechanism underlying this response might involve ATM, a central transducer in the DDR, because ATM-deficient cells fail to regulate interferon during the response to genotoxic stress (Pamment et al., 2002). However, these ATM-deficient cells can still activate interferon in response to stimuli other than DNA damage. Accordingly, ATM is not only present in the nucleus, but also in the cytoplasm (Hinz et al., 2010). It is possible that the DDR-dependent interferon activation has emerged as an evolutive response to DNA damage induced by viruses, working as a mechanism that reduces cell proliferation to allow DNA repair or promote cell death.

Whereas the studies mentioned so far reveal a direct link between innate immune signaling and the response of cells to induced DNA damage, aging could radically affect the scenario. Dysfunctional telomeres, DNA damage, and the persistent response to these events eventually trigger cellular senescence, a state of irreversible cell cycle arrest. Cells bearing senescent markers increase with age in a variety of tissues in mice (Krishnamurthy et al., 2004; Wang et al., 2009) and in primates (Herbig et al., 2006; Jeyapalan et al., 2007). Although senescence was initially understood as a protective mechanism to suppress the development of cancer and promote tissue repair, this cellular mechanism is now seen as a double-edged sword (Campisi, 2011). A role for senescence in tissue repair would probably explain the evolution of the
so-called senescence-associated secretory phenotype (SASP) (Coppé et al., 2008). The SASP entails the secretion of factors, such as the proinflammatory cytokines IL (interleukin)-6 and IL-8. Although there is evidence that the SASP suppresses tumor formation by reinforcing cellular senescence, it also promotes cancer progression by stimulating the growth of nearby precancerous cells. The most convincing evidence for this activity comes from xenograft studies. Co-injection of senescent fibroblasts significantly stimulated the proliferation of mouse and human epithelial tumor cells, while co-injection of nonsenescent fibroblasts did not (Liu & Hornsby, 2007). As cancers are among the pathologies that are fueled by inflammation (Grivennikov et al., 2010), the cytokines that comprise the SASP in aging organisms, together with the innate immune responses triggered by DNA damage, can synergistically contribute to age-related cancer by stimulating inflammation.

**Nuclear structure and radiosensitivity**

**Aging-related changes in the nuclear organization hinder proper DNA Repair**

Age-related changes have been detected in the nuclear lamina and the nuclear pore complexes, the main components of the peripheral nuclear matrix. These changes can lead to chromatin reorganization and alterations in the epigenetic status. Recent studies have highlighted the relevance of the lamina in the maintenance of the nuclear architecture and genome integrity and have shown that age-related changes induce poor DNA repair efficiency and fidelity.

Lamins are type V intermediate filaments located just inside the nuclear inner membrane, forming a meshwork throughout the nucleoplasm called nuclear lamina. Two splice variants of A-type lamins, lamin A, and lamin C, together with lamin B, participate in many essential nuclear processes. Among them, lamins contribute to DNA replication and repair (Goldman et al., 2004; Oberdoerffer & Sinclair, 2007; Mewborn et al., 2010; Camps et al., 2014). The relationship between lamins and aging has been approached through the study of Hutchinson–Gilford progeria syndrome (HGPS), a rare condition of premature aging caused by a mutation in LMNA, the gene encoding A-type lamins. These mutations translate to the accumulation of an immature nonfunctional protein (progerin) that causes changes in gene expression, heterochromatin organization, and failure of proper DNA repair (Liu et al., 2005). Interestingly, the accumulation of the immature prelamin A has also been reported in normal dermal fibroblasts from old individuals (Scaffidi & Misteli, 2006), in *in vitro* senescent human fibroblasts (Cao et al., 2007), and in aged vascular smooth muscle cells (Ragnauth et al., 2010). Therefore, production of progerin in normal aging cells may be inducing similar phenotypic features to those reported in laminopathies such as defective DNA repair, changes in heterochromatin organization, and telomere attrition (Gonzalez-Suarez & Gonzalo, 2010).

In fact, several studies unmasked the close relationship between telomere attrition, nuclear integrity, and DNA repair. Cao and colleagues demonstrated that progressive telomere attrition in normal cells acts as an upstream signal to activate the cryptic splice site in LMNA to produce progerin. They also suggested that progerin induces an acute DNA damage response at telomeres that leads to depletion of the telomeric 3’ overhang (Cao et al., 2011). In line with this, fibroblasts from LMNA null mouse model showed an altered telomere structure and function together with compromised efficiency of NHEJ and HR pathways, as recruitment of 53BP1 and RAD51 to DSBs was delayed in progeria cells (Liu et al., 2005; Gonzalez-Suarez et al., 2009). Moreover, it has been suggested that compromising the expression of lamin A causes 53BP1 degradation, as A-type lamins seem to be involved in the stabilization of 53BP1, preventing its degradation by the proteasome (Gonzalez-Suarez & Gonzalez, 2010). The loss of lamin proteins also leads to a defective HR pathway, due to the formation of p130/E2F4 complexes, which bind RAD51 and *BRCA1* promoters and inhibit their transcription (Haithcock et al., 2005; Redwood et al., 2011).

![Fig. 4](image-url)  
**Fig. 4** Scheme of the age-related defects in nuclear reorganization. The aging process entails changes at the nuclear organization level that may compromise the DNA damage response (DDR) proteins recruitment to the nucleus: accumulation of a premature and dysfunctional prelamin A, untethering of the heterochromatin (HC) domains, and an increased presence of leaking pores. The addition of radiation by means of reactive oxygen species (ROS) production hinders both NPC and lamins function by oxidation.
Therefore, lamins deficiency or mutation can induce unprotected telomeres and compromise the accumulation of DNA repair proteins, resulting in impaired DSB repair (Liu et al., 2005; Redwood et al., 2011). Because normal aging cells can accumulate progerin, radiation exposure and/or oxidative stress may induce genomic instability among the elderly.

Nuclear pore complexes (NPCs) can also contribute to the DDR deterioration. Dysfunction of the nuclear pore complexes can result in the loss of essential proteins to maintain the chromatin organization and DNA repair. NPCs work as nuclear gatekeepers, allowing the free diffusion of small molecules and tightly regulating the transport of macromolecules (Fernandez-Martinez & Rout, 2009). The lifespan of some scaffold nucleoporins is exceptionally long (D’Angelo et al., 2009), and thus, the lack of turnover contributes to NPCs deterioration by age-related oxidative stress (Savas et al., 2012). Importantly, damaged NPCs are more permeable, which causes leaking of cytoplasmic proteins, such as tubulin, into the nucleus (D’Angelo et al., 2009). Permeable NPCs could also disturb the nuclear presence of DDR factors leading to an inefficient DNA repair. Altogether, aging may compromise several roles of the nuclear envelope putting the genome integrity at risk.

Ionizing radiation corrodes the already disturbed nuclear organization in the aging cells

Age-related changes described above, such as nuclear lamina decay and leaking NPCs, hamper the proper repair of radiation-induced DNA lesions. Hence, the increased radiosensitivity observed in aging cells may be a consequence of already dysfunctional systems trying to cope with the damage raised by IR. As stated before, ionizing radiation does not only induce DNA lesions, but also disturbs the nuclear organization by ROS production (Fig. 4). Indeed, the lamina and the NPC might be functionally modified by exposure to free radicals that accumulate in aging cells and that are derived from IR exposure. In this line, increased levels of carboxyl groups, indicative of oxidative protein damage, have been found in those nucleoporins forming age-related damaged leaky NPCs (D’Angelo et al., 2009). When the nuclear lamina is exposed to chronic or acute oxidative stress, irreversible damage is induced to their conserved cysteine residues, impeding their function (Pekovic et al., 2011; Sieprath et al., 2012). In light of these results, it is plausible to hypothesize that irradiating aged cells would promote further protein oxidation and, as a consequence, further nuclear disorganization hampering the repair of radiation-induced DNA lesions and thus jeopardizing the maintenance of genome integrity.

Conclusions

There are many factors that contribute to the close relationship that exists between radiation sensitivity and age. These factors provide several plausible and nonexclusive explanations for the increased carcinogenic risk of radiation in the elderly, as reported by epidemiological studies in different cohorts. It is possible that the age-related oxidative stress accumulation disturbs the nuclear organization, which in turn may impede a proper repair. Because nuclear organization is also disturbed by ionizing radiation, aging cells become an easy target for suffering enhanced effects after irradiation. DDR impairment together with telomere attrition can result in the accumulation of genomic rearrangements that may contribute to the increased incidence of radiation-induced carcinogenesis in the elderly. Nevertheless, the intricate interplay between these aging-related defects acts as a confounding factor making it very difficult to study the contribution of each factor independently. Thus, studies that provide an integrated view of how radiosensitivity affects aging organisms are necessary for a better understanding and improvement of experimental designs. We conclude that aging must be seen as a convoluted scenario, where radiation is a bump in the road of preserving genome integrity. Because radiosensitivity increases with age and life expectancy rises steadily, there is a need for improving radiation protection measures to ensure the safety of the elderly.

Acknowledgments

The authors would like to apologise to those whose work has not been cited due to space constraints. The authors thank the Language Advisory and Translation Unit at Universitat Autònoma de Barcelona Language Service for editing the manuscript.

Funding

This work was supported by grants from Consejo de Seguridad Nuclear (CSN 2012-0001) and EURATOM (Dark.Risk GA 323216) to AG. The authors acknowledge 2014-SGR-524 grant from Generalitat de Catalunya. JC is supported by the Asociacion Española Contra el Cancer and the Instituto de Salud Carlos III (CP13/00160). LT acknowledges Ministerio de Economia y Competitividad grant (SAF2013-43801-P).

Conflicts of interest

The authors declare that have no conflict of interests.

References

d’Adda di Fagagna F, Reapers PM, Clay-Farrance L, Fiegler H, Carr P, Von Zglinicki T, Saretzki G, Carter NP, Jackson SP (2003) A DNA damage checkpoint response in telomere-initiated senescence. Nature 426, 194–198.

Andersen HR, Nielsen JB, Nielsen F, Grandjean P (1997) Antioxidative enzyme activities in human erythrocytes. Clin. Chem. 43, 562–568.

Artandi SE, Chang S, Lee SL, Alston S, Gottlieb GJ, Chin L, DePinho RA (2000) Telomere dysfunction promotes non-reciprocal translocations and epithelial cancers in mice. Nature 406, 641–645.

Bekker-Jensen S, Mailand N (2010) Assembly and function of DNA double-strand break repair foci in mammalian cells. DNA Repair (Amst). 9, 1219–1228.

Brouillet S, Singh RK, Thompson JR, Goodall AH, Samani NJ (2003) White cell telomere length and risk of premature myocardial infarction. Arterioscler. Thromb. Vasc. Biol. 23, 842–846.

Brouillet SW, Moore JS, McMahon AD, Thompson JR, Ford I, Shepherd J, Packard CJ, Samani NJ (2007) Telomere length, risk of coronary heart disease, and statin treatment in the West of Scotland Primary Prevention Study: a nested case-control study. Lancet 369, 107–114.

Brzostek-Racine S1, Gordon C, Van Scoy S, Reich NC (2011) The DNA damage response induces IFN. J. Immunol. 187, 5336–5345. doi: 10.4049/jimmunol.1100040. Epub 2011 Oct 17.

Buffenstein R, Edrey YH, Yang T, Mele J (2008) The oxidative stress theory of aging: embattled or invincible? Insights from non-traditional model organisms. Age (Dordr) 30, 99–109.

Burhans WC, Weinberger M (2007) DNA replication stress, genome instability and aging. Nucleic Acids Res. 35, 7545–7555.

Campisi J (2011) Cellular senescence: putting the paradoxes in perspective. Curr. Opin. Genet. Dev. 21, 107–112.

Campis J, Wangsa D, Falke M, Brown M, Case CM, Erd MR, Ried T (2014) Loss of lamin B1 results in prolongation of S phase and decondensation of chromosome territories. FASEB J. (2014) 28, 3423–3434. [Epub ahead of print].

Cao K, Capell BC, Erdos MR, Djabali K, Collins FS (2007) A lamin A protein isoform overexpressed in Hutchinson-Gilford progeria syndrome interferes with mitosis in progenitor and normal cells. Proc. Natl Acad. Sci. USA 104, 4949–4954.

Cao K, Blair CD, Faddah DA, Kiekhaefer JE, Olive M, Erdos MR, Nabel EG, Collins FS (2011) Progerin and telomere dysfunction collaborate to trigger cellular senescence in normal human fibroblasts. J. Clin. Invest. 121, 2833–2844.
Chowdhury D, Choi YE, Brault ME (2013) Charity begins at home: non-coding RNA functions in DNA repair. Nat. Rev. Mol. Cell Biol. 14, 181–189.

Colgin LM, Redick TR (2009) Telomere maintenance mechanisms and cellular immortalization. Curr. Opin. Genet. Dev. 19, 97–103.

Coppé JP, Patil CK, Rodier F, Sun Y, Murzio DP, Goldstein J, Nelson PS, Desprez PY, Campisi J (2008) Senescence-associated secretory phenotypes reveal cell-nonautonomous functions of oncogenic RAS and the p53 tumor suppressor. PLoS Biol. 6, 2853–2868.

D’Angelo MA, Raices M, Panovski SH, Hetzer MW (2009) Age-dependent deterioration of nuclear pore complexes causes a loss of nuclear integrity in postmitotic cells. Cell 136, 284–295.

De Magalhães JP (2013) How aging processes influence cancer. Nat. Rev. Cancer 13, 357–365.

Fang X, Yoon J-G, Li L, Yu W, Shao J, Hua D, Zheng S, Hood L, Goodlett DR, Foltz G, Lin B (2011) The SOX2 response program in glioblastoma multiforme: an integrated ChIP-seq, expression microarray, and microRNA analysis. BMC Genom. 12, 11.

Fazel R, Krumholz HM, Wang Y, Ross JS, Chen J, Ting HH, Shah ND, Nasir K, Fitzpatrick AL, Kronmal RA, Gardner JP, Psaty BM, Jenny NS, Tracy RP, Walston J, Curr. Fordel E, Thijs L, Martinet W, Lenjou M, Laufs T, Van Bockstaele D, Moens L, Goldman RD, Shumaker DK, Erdos MR, Eriksson M, Goldman AE, Gordon LB, ~ Haithcock E, Dayani Y, Neufeld E, Zahand AJ, Feinstein N, Mattout A, Gruenbaum Hemmi H, Takeuchi O, Kaisho T, Sato S, Sanjo H, Matsumoto M, Hoshino Mutat. Res. Harman D (1992) Free radical theory of aging. Aging and radiation, L. Hernández Copp Grivennikov SI, Greten FR, Karin M (2010) Immunity, inflammation, and cancer.

Inal ME, Karbassi G, Sunal E (2001) Antioxidant enzyme activities and malondialdehyde levels related to aging. Am. J. Epidemiol. 165, 14–21.

Foord E, Thijs L, Martinet W, Lenjou M, Laufts T, Van Bockstaele D, Moens L, Dewilde S (2006) Neuroglobin and cytoglobin overexpression protects human SH-SY5Y neuroblastoma cells against oxidative stress-induced cell death. Neurosci. Lett. 410, 146–151.

Goldman RD, Shumaker DK, Erods MR, Eriksson M, Goldman AE, Gordon LB, Gruenbaum Y, Khoun S, Mendez V, Varga R, Collins FS (2004) Accumulation of mutant lamin A causes progressive changes in nuclear architecture in Hutchinson-Gilford progeria syndrome. Proc. Natl Acad. Sci. USA 101, 8963–8968.

Fernandez-Martinez J, Rout MP (2009) Nuclear pore complex biogenesis. Curr. Opin. Cell Biol. 21, 603–612.

Fitzpatrick AL, Kronmal RA, Gardner JP, Psaty BM, Jenny NS, Tracy RP, Waldston J, Curr. Einstein AJ, Nallamothu BK (2009) Exposure to low-dose ionizing radiation from increased tumor incidence in older populations. Declining p53 function in the aging process: a possible mechanism for the increased tumor incidence in older populations. Proc. Natl Acad. Sci. USA 104, 16633–16638.

Goytisolo FA, Samper E, Martinez-Cinca JC, Pinos E, Herrera E, Flores JM, Bouffler SV, Kupke M, Kanbak G, Sunal E (2001) Antioxidant enzyme activities and malondialdehyde levels related to aging. Am. J. Epidemiol. 165, 14–21.

Joyce NC, Harris DL, Zhu CC (2011) Age-related gene response of human corneal epithelial cells aged 461, 1071–1078.

Jeyaraj IC, Ferreira M, Sedivy JM, Herbig U (2007) Accumulation of senescent cells in mitotic tissue of aging primates. Mech. Aging Dev. 128, 36–44.

Joyce NC, Harris DL, Zhu CC (2011) Age-related gene response of human corneal endothelium to oxidative stress and DNA damage. Invest. Ophthalmol. Vis. Sci. 52, 1641–1649.

Junqueira VBC, Barros SBM, Chan SS, Rodrigues L, Giavartoli A, Abud RL, Deucher GP (2004) Aging and oxidative stress. Mol. Aspects Med. 25, 5–16.

Karakasilioti I, Kamileri I, Chatzinikolaou G, Kosteas T, Vergadi E, Robinson AR, Tse L, Ezan F, Song XP, Liu Y, Sun Y, Mu SC, Zick D, Ono K, Sharpless NE (2004) Ink4a/Arf expression is a biomarker of aging. J. Clin. Invest. 114, 1299–1307.

Kreigl K, Zhang H (2007) An integrated view of oxidative stress in aging: basic mechanisms, functional effects, and pathological considerations. Am. J. Physiol. 382, R18–R36.

Kinchin namurthy J, Torrice C, Ramsey MR, Kovalev GI, Al-Regaiey K, Su L, Sharpless NE (2004) Ink4a/Arf expression is a biomarker of aging. J. Clin. Invest. 114, 1299–1307.

Ku HH, Brunk UT, Sohal RS (1993) Relationship between mitochondrial superoxide and hydrogen peroxide production and longevity of mammalian species. Free Radic. Biol. Med. 15, 621–627.

Lapointe J, Heikni S (2010) When a theory of aging ages badly. Cell. Mol. Life Sci. 67, 1–8.

Latre L, Tussell L, Martin M, Miró R, Egozcue J, Blasco M, Genescà A (2003) Shortened telomeres join to DNA breaks interfering with their correct repair. Exp. Cell Res. 287, 282–288.

Li L, Zhang L, Fan J, Greenberg K, Desiderio S, Rassool PV, Small DA (2011) Defective nonhomologous end joining and blocks B-cell development in FLT3/ITD mice. Blood 117, 3313–3319.

Lieber MR (2008) The mechanism of human nonhomologous DNA end joining. J. Biol. Chem. 283, 1–5.

Liu D, Hornsby JP (2007) Senescent human fibroblasts increase the early growth of xenograft tumors via matrix metalloproteinase secretion. Cancer Res. 67, 3117–3126.

Liu D, Xu Y (2011) p53, Oxidative stress, and aging. Antioxid Redox Signal. 15, 1669–1678.

Liu B, Wang J, Chen KM, Tija WM, Deng W, Guan X, Huang J, Li KM, Chau PY, Chen DJ, Pei D, Pendas AM, Caballero J, López-Otin C, Tse HF, Hutchison C, Chen J, Cao Y, Cheah KSE, Tryggvason K, Zhou Z (2005) Genomic instability in laminopathy-based premature aging. Nat. Med. 11, 780–785.

López-Otin C, Blasco MA, Partridge L, Serrano M, Kroemer G (2013) The hallmarks of aging. Cell 153, 1194–1217.

Lushnikova T, Bouska A, Odvody J (2011) Aging mice have increased chromosome instability that is exacerbated by elevated Mdm2 expression. Oncogene 30, 4622–4631.

Mao J, Tian X, Van Meter M, Ke Z, Gorbunova V, Seluanov A (2012) Sirtuin 6 (SIRT6) rescues the decline of homologous recombination repair during replicative senescence. PNAS 11, 11800–11805.

Meeker A, Hicks J (2004) Telomere length abnormalities occur early in the initiation of epithelial carcinogenesis. Clin Cancer Res. 10, 3317–3326.

Mewborn SK, Puckewitz MJ, Abusunnein FH, Fahrenbach JP, Zhang Y, MacLeod H, Dellefave L, Pytel P, Selig S, Caballero J, Reddy K, Singh H, McNally E (2010) Altered chromosomal positioning, compaction, and gene expression with a lamin A/C gene mutation. Mol. Cell 37, 780–785.

Negrini S, Gorgoulis VG, Halazonetis TD (2010) Genomic instability–an evolving hallmark of cancer. Nat. Rev. Mol. Cell Biol. 11, 220–228.

Oberdoerffer P, Sinclair DA (2007) The role of nuclear architecture in genomic stability and aging. Nat. Rev. Mol. Cell Biol. 8, 692–702.

Pamment J, Ramsay E, Kelleher M, Dornan D, Ball KL (2002) Regulation of the IRF-1 tumour modifier during the response to genotoxic stress involves an ATM-dependent signalling pathway. Oncogene 21, 7776–7785.

Pekovic V, Gibbs-Seymour I, Markiewicz E, Alzoghaibi F, Benham AM, Edwards R, Wenhert M, von Zglinicki T, Hutchinson CJ (2011) Conserved cysteine residues in the mammalian lamin A tail are essential for cellular responses to ROS generation. Aging Cell 10, 1067–1079.
Perez VI, Bokov A, Van Remmen H, Mele J, Ran Q, Ikeno Y, Richardson A (2009) Is the oxidative stress theory of aging dead? Biochim. Biophys. Acta 1790, 1005–1014.

Puthiyaveetil AG, Caudell DL (2013) Non homologous end joining-mediated DNA break repair is impaired in B lymphocytes of aging mice. Mol. Immunol. 53, 79–87.

Ragagnin CD, Warren DT, Liu Y, McNair R, Tajsc T, Figg N, Shroff R, Skepper J, Shanahan CM (2010) Prelamin A acts to accelerate smooth muscle cell senescence and is a novel biomarker of human vascular aging. Circulation 121, 2200–2210.

Redwood AB, Perkins SM, Vanderwaal RP, Feng Z, Biehl KJ, Gonzalez-Suarez I, Morgado-Palacin L, Shi W, Sage J, Roti-Roti JL, Stewart CI, Zhang J, Gonzalez S (2011) A dual role for A-type lamins in DNA double-strand break repair. Cell Cycle 10, 2549–2560.

Richardson RB (2013) P53 mutations associated with aging-related rise in cancer incidence rates. Cell Cycle 12, 2468–2478.

Richardson DB, Wing S (1999) Greater sensitivity to ionizing radiation at older age: follow-up of workers at Oak Ridge National Laboratory through 1990. Int J Epidemiol. (1999) 28, 428–436.

Riley PA (1994) Free radicals in biology: oxidative stress and the effects of ionizing radiation. Int. J Radiat. Biol. 65, 27–33.

Rogakou E, Pilch D, Orr A (1998) DNA double-stranded breaks induce histone H2AX phosphorylation on serine 139. J. Biol. Chem. 273, 5858–5868.

Rubé CE, Frick A, Widmann TA, Frst T, Madry H, Pfreundschuh M, Rubé C (2011) Accumulation of DNA damage in hematopoietic stem and progenitor cells during human aging. PLoS One. (2011) 6, e17487.

San Filippo J, Sung P, Klein H (2008) Mechanism of eukaryotic homologous recombination. Annu. Rev. Biochem. 77, 229–257.

Savas JN, Toyama BH, Xu T, Yates JR, Hetzer MW (2012) Extremely long-lived nuclear pore proteins in the rat brain. Science 335, 942.

Scaffidi P, Misteli T (2006) Lamin A-dependent nuclear defects in human aging. Annu. Rev. Biochem. 77, 11–21.

Seluanov A, Mittelman D, Pereira-Smith OM, Wilson JH, Gorbunova V (2004) DNA end joining becomes less efficient and more error-prone during cellular senescence. PNAS 101, 7624–7629.

Seluanov A, Danek J, Hause N, Gorbunova V (2007) Changes in the level and distribution of Ku proteins during cellular senescence. DNA Repair (Amst). 6, 1740–1748.

Seong KM, Kim CS, Jeon HY, Oh S-H, Nam SY, Yang KH, Kim J-Y, Jin Y-W (2010) Intrinsic radiosensitivity correlated with radiation-induced ROS and cell cycle regulation. Mol. Cell. Toxicol. 6, 1–7.

Shuryak I, Sachs RK, Brenner DJ (2010) Cancer risks after radiation exposure in middle age. J. Natl Cancer Inst. 102, 1628–1636.

Separth T, Darwiche R, De Vos WH (2012) Lamins as mediators of oxidative stress. Biochem. Biophys. Res. Commun. 421, 635–639.

Soler D, Genescá A, Arnedo G, Egózcue J, Tusell L (2005) Telomere dysfunction drives chromosomal instability in human mammary epithelial cells. Genes Chromosom. Cancer 44, 339–350.

Sosa V, Moliné T, Somoza R, Pacucci R, Kondoh H, Lleonart ME (2013) Oxidative stress and cancer: an overview. Ageing Res. Rev. 12, 376–390.

Stetson DB, Medzhitov R (2006) Recognition of cytosolic DNA activates an IRF3-dependent innate immune response. Immunity 24, 93–103.

Tanaka H, Abe S, Huda N, Tu L, Beam MJ, Grimes B, Gilley D (2012) Telomere fusions in early human breast carcinoma. Proc. Natl Acad. Sci. USA 109, 14098–14103.

Valio M, Leibfritz D, Moncol J, Cronin MTD, Mazur M, Telser I (2007) Free radicals and antioxidants in normal physiological functions and human disease. Int. J. Biochem. Cell Biol. 39, 44–84.

Veskoukis AS, Tsatsakis AM, Kouretas D (2012) Dietary oxidative stress and antioxidant defense with an emphasis on plant extract administration. Cell Stress Chaperones. 17, 11–21.

Wang C, Jurk D, Maddick M, Nelson G, Martin-Ruiz C, von Zglinicki T (2009) DNA damage response and cellular senescence in tissues of aging mice. Aging Cell 8, 311–323.

Weiss JP, Landauer MR (2003) Protection against ionizing radiation by antioxidant nutrients and phytochemicals. Toxicology 189, 1–20.

Wong KK, Chang S, Weiler SR, Ganesan S, Chaudhuri J, Zhu C, Artandi SE, Wong KK, Chang S, Weiler SR, Ganesan S, Chaudhuri J, Zhu C, Artandi SE, Rudolph KL, Gottlieb GJ, Chin L, Alt FW, DePinho R a (2000) Telomere dysfunction impairs DNA repair and enhances sensitivity to ionizing radiation. Nat. Genet. 25, 85–88.

Wyman C, Kanaar R (2006) DNA double-strand break repair: all’s well that ends well. Annu. Rev. Genet. 40, 363–383.