Review Article
The diverse and complex roles of radiation on cancer treatment: therapeutic target and genome maintenance

Rajamanickam Baskar[1,2], Swee Peng Yap[1], Kevin Lee Min Chua[1], Koji Itahana[3]

[1]Department of Radiation Oncology; [2]Division of Cellular and Molecular Research, National Cancer Centre; [3]Cancer and Stem Cell Biology Program, Duke-NUS Graduate Medical School, Singapore

Abstract: Cancer is a genetic disease, grows exponentially with the development of intrinsic and acquired treatment resistance. Past decade has witnessed a considerable progress towards the treatment and understanding of proposed hallmarks of cancer and together with advances in early detection and various treatment modalities. Radiation therapy is an integral part of cancer treatment armamentarium. In developed countries more than half of all cancer patients receive radiation therapy during their course of illness. Although radiation damages both cancer and normal cells, the goal of radiation therapy is to maximize the radiation dose to abnormal cancer cells while minimizing exposure to normal cells, which is adjacent to cancer cells or in the path of radiation. In recent years, life expectancy increases among cancer patients and this increase is due to the results of early diagnosis, screening efforts, improved treatments and with less late effects mostly secondary cancer development. Therefore, cancer survivorship issues have been gaining prominence in the area of radiation oncology research. Understanding the tradeoff between the expected decreases in normal tissue toxicity resulting from an improved radiation dose distribution to the targeted site is an increasingly pertinent, yet needed attention and research in the area of radiation oncology. In recent years, a number of potential molecular targets that involve either with radiation increased tumor cell killing or protecting normal cells have been identified. For clinical benefits, translating these findings to maximize the toxicity of radiation on tumor cells while safeguarding early or late normal cell toxicities using molecular targeted radioprotectors will be useful in radiation treatment.

Keywords: Cancer, radiation therapy, radioprotectors, normal genome maintenance

Introduction

Despite decreases in cancer related death rates in developed countries like USA and in western countries, the number of cancer cases and deaths are projected to be more than double worldwide in the next 20-40 years [1, 2]. The projected increase will be driven largely by growth and aging of populations. Past decade has witnessed a major leap in the understanding of molecular mechanisms involved in tumor pathogenesis, progression and further identified various treatment modalities to control this complex disease. Despite initial high response rates to the various treatment modalities and interventions, a large proportion of cancer patients suffered relapse in years or decades later [3-6], resulting a therapeutic challenge. International Agency for Research on Cancer (IARC) has predicted that by 2030, 27 million new cancer

cases and 17 million cancer deaths will occur each year worldwide. That compares to 12.7 million new cancers and 7.6 million cancer death reported by GLOBOCAN 2008 [7].

Cancers are primarily an environmental disease with 90-95% of cases are due to modification in lifestyle and environmental factors, and only 5-10% of cancers occur due to an abnormality inherited from mother or father [8, 9]. Thus, cancer is largely considered a preventable disease. In recent years, many treatment and management options for cancer exist with the primary ones including: surgery, chemotherapy, radiotherapy and palliative care.

Radiation is an invaluable diagnostic and treatment tool used in various clinical applications. Radiation therapy or radiotherapy is a cornerstone of modern cancer management is a highly

effective and widely used to destroy (kill) cancer cells. More than half of all cancer patients mostly in the developed countries receive radiation in the form radiotherapy using various radiation sources [10-12] to cure the disease either alone or in combination with other treatment modalities such us chemotherapy or surgery. Radiotherapy is a highly cost effective single modality treatment, accounting about only 5% of the total cost of cancer care [13]. Radiotherapy (external beam or internal irradiation given as brachytherapy: such as protons, heavy ions, as well as a largely used standard sources –photons) is the most important non-surgical modality for the curative treatment of cancer, but its curative potential is often limited by intrinsic radioresistance of cancer cells/mass and normal cell toxicity.

The first clinical use of radiation for the cancer treatment was recorded in late 19th century [14, 15], soon after Roentgen described X-rays in 1895 and the effectiveness of radiotherapy that has been developed over the years showed an increase in the number of cancer survivors. As more cancer patients undergo radiotherapy and live longer after treatment, the number of cancer survivors in the United States America (USA) has tripled since 1971 and is growing by 2% each year [16, 17], therefore radiotherapy benefit a large number of cancer patients. Though accurate delivery of the radiation dose has been greatly improved, nonuniform dose distributions in the adjacent normal tissues or rests of patient body is inevitable and further this effect also mediated by cellular and tumor microenvironmental signaling [6, 18, 19]. Furthermore, radiation-induced damage of normal tissues surrounding the tumors limits the effectiveness of radiation treatment [20-22]. Long term post radiation exposure survivors particularly children face various health effects and also increased risk of post treatment cancer development in adjacent or at distanced organs, risk of second cancers or late toxicities [23-25]. However, advances in pediatric cancer treatment techniques and care yielded increase in survival during the last four decades. In the USA, approximately 90.4% of children with cancer will survive 5 years or longer in recent years (2000-2005) compared with 83.7% in 1990-1994 [24]. Therefore, there is an increasing concern regarding the radiation related risks or toxicities in long-term radiotherapy survivors [26-29] and counteracting these effects to achieve clinical efficacy.

Radiation and cell's response

Radiotherapy is based on the fact that ionizing radiation (a physical agent) can induce damage in biological tissues and thus efficiently kill tumor cells. In radiation oncology high-energy of radiation (typically 1 MeV or more) is used to eradicate the proliferating cancer cells. Biological effectiveness of radiation depends on the linear energy transfer (LET) that penetrate and deposits energy in tissues, total dose, fractionation rate and radio-sensitivity of the targeted cells or tissues. For cancer treatment, external beam radiotherapy is primarily used for local tumor irradiation. Radiotherapy can also be delivered in the form of targeted radionuclides (radiation is attached to a 'carrier' that selectively kills cancer cells by delivering a lethal dose of radiation) or brachytherapy (placement of radioactive sources in or just next to the tumor). Irradiation is most commonly delivered via photons released from a linear accelerators (X-rays, gamma rays and beta particles; low LET radiations), deposits a relatively small quantity of energy on the targeted tissue. On the other hand, high LET radiations protons (from a cyclotron or synchrotron) deposits more energy on the targeted areas and further causes more biological effects than the low LET radiations. Radiation damages the genetic material (DNA) causing single or double strand breaks, DNA base damage, DNA-protein cross links with chromosomal rearrangements in the cells and thus blocking their ability to divide and proliferate further [30, 31] (**Figure 1**), all these incidences are proportional to the absorbed dose. Although radiation damages both cancer cells as well as normal cells, the advancement in 3D conformal radiotherapy, intensity-modulated radiotherapy and proton beam therapy have all been developed to maximize the radiation dose to abnormal cancer cells while sparing the normal surrounding cells, preventing or reducing normal cells death or late effects has increasingly become a priority in the field of radiation oncology. A complementary approach is to explore mechanisms of protecting or reducing the normal tissue injury while increase the tumor cell killing probability using molecular targeted radioprotectors, which protect the genetic nature of normal cells are evolving. Although we do not have a comprehensive answer about the molecular mechanism of radiation-induced cancer on individual variations in susceptibility, especially of therapy-related cancers, it is well known that cancer is a complex multistep process and ra-
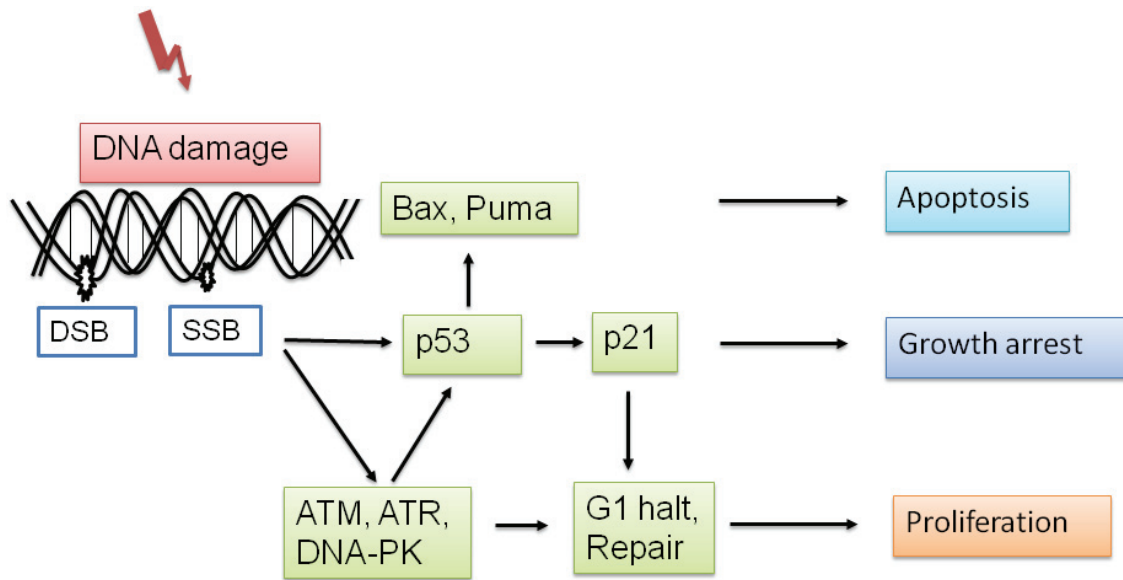
Figure 1. Radiation damages the genetic material (DNA) causing single strand breaks (SSB) or double strand breaks (DSB) in the cells, thus blocking their ability to divide and proliferate further.

diation can influence both initiation and promotion. During the past decade our knowledge of molecular basis of cancer cell resistance and survival has changed our understanding to maximize therapeutic benefits of radiation. Therefore, understanding this problem at a molecular level and any improvement in the efficacy of radiotherapy treatment will benefit a large number of cancer patients.

Radiation is administered single or mostly fractionated with small doses (1.8-2 Gy) of radiation over a period of 5-7 weeks [32]. Furthermore fractionated radiotherapy schedule will allow the recovery of normal cells from sub-lethal damage between radiation schedules, which provides enough time to repopulate the normal cells. Although repopulation also occurs in cancer cells, rapid cell proliferation or accelerated doubling time of cancer cells in which "key signals for growth and cell divisions are always on" than the normal cells [32, 33] showed more radiosensitivity for the better tumor control. In normal and cancer cells, radiation mainly causes accumulation of DNA either directly or by indirectly (through free-radicals) interfering with DNA synthesis is the common lethal mechanisms involved [34]. Cancer cells whose DNA is damaged beyond repair stop dividing and die. X-ray dose of ~1 Gy produces about 1000 single strand breaks (SSB) and about 50-

100 double strand breaks (DSB) in a typical mammalian cell [35]. Although both single and double strand DNA breaks are observed, DSBs are the most responsible factor involved in radiation induced cell death [36]. Cells with DNA lesions (for example, incomplete replication and hypomethylation) arrest in the cell cycle at either the G_1-S or the G_2-M transition (G_1 and G_2 checkpoints, respectively). This allows the DNA repair machinery time to attempt repair it's genome and retain the genomic integrity. If the degree of DNA damage is beyond recovery, cells never enter mitosis, stop dividing and undergo cell death (also known as 'mitotic catastrophe'). Radiotherapy does not kill cancer cells right away. It takes hours, days or weeks of treatment before cancer cells start to die. Thus, identifying the importance of radiation induced effective cancer cell deaths and further mechanisms involved in protection of normal cells especially fast growing lymphoid organs, bone marrow, intestinal crypts, testes, and ovaries has potential and clinical implications for improving outcomes with radiotherapy.

Protective nature of cancer cells

Genetic and molecular approaches have fostered remarkable progress in our understanding of cancer development and also control/cure this growing disease. In the process of tumor
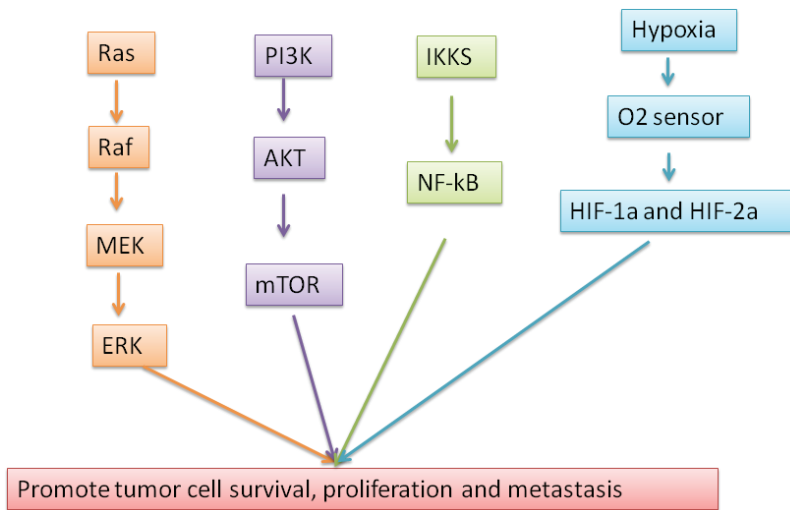
Figure 2. Cytoprotective and prosurvival proteins/pathways involved in tumor cell survival, proliferation and metastasis.

formation, cancer cells acquired cytoprotective and prosurvival proteins/pathways (**Figure 2**) during the transformation process, suggesting that these pathways are important for tumor development and growth. Radiotherapy, like most anticancer treatments, achieves its therapeutic effect by interfering cell's cell cycle by halting the cell cycle and ultimately inducing cell death. However, various DNA repair mechanisms within the cancer cells interfere with the radiation induced damage and further increase the radioresistance of cancer cells [37]. Besides these repair pathways, signal transduction pathways are also involved in cellular response to radiation and this cellular responses showed an important implications for fractionated radiotherapy for the cancer cell survival, a protectant mechanisms of cancer cells for any genotoxic insults or radioresistant of the cancer cells is a major clinical problem. Of the many signaling cascades governing the stress response of a cell to various internal and external stimuli, it has been shown that four pathways demonstrated a clear role on the resistance to anti cancer treatment. Three of these PI3K-AKT-mTOR, nuclear factor -kB (NF-kB) and MAPK are involved in cell survival pathways (**Figure 2**) and further DNA damage activates ATM /ATR (ataxia telangiectasia-mutated/ ataxia telangiectasia and Rad3-related) which, in turn, activates proteins involved in cell-cycle control and DNA repair. Depending on the extent of damage and the repair capability of cells, the outcome is

either cancer cell death or survival. Among these pathways, PI3K-AKT-mTOR signaling pathway is regarded as most important for the cancer cell survival and proliferation [38, 39], and further involved in the resistance to the anti-cancer treatment [40].

Oxygen is an essential regulator of cellular metabolism, survival and cell proliferation. Solid cancers are heterogeneous, in addition solid cancers also express hypoxic regions due to high rates of cell proliferation coupled with the formation of structurally and functionally poor vasculature with less nutrition support to divide and grow [41-43]. It has been shown that increased levels of hypoxia-inducible factors (HIF-1a or HIF-2a) in diagnostic tumor biopsies are associated with increased risk of mortality in cancers of bladder, brain, breast, colon, cervix, endometrium, head/neck, lung, ovary, pancreas, prostate, rectum, and stomach [44]. HIFs is a transcription factor which activates various genes that play key role in cancer progression/metastasis [45], including radiation resistance [46], with a potentially promising target for tumor radiosensitisation. Thus counteracting these stress response pathways will be toxic to the cancer cells, prevent further spreading into other part of the body and eventually kill the cancer cells. Defects in these cell proliferation pathways are hallmarks of cancer development [47, 48] and further radiotheraphy alone or with chemotherphy can remove the cancer cells and prevent the cancer spreading.

Normal cell radioprotection and challenges

In recent decades accurate delivery of radiation to the tumor site has greatly improved. However, it is not always feasible to eliminate a tumor without damaging the surrounding normal tissue. Tumor cells heavily rely on the activation of one or two pathways, a phenomenon known as oncogene addiction [49, 50], whereas normal cells use a broader range of molecular signals to overcome various cellular insults [51] to maintain the normal genome. Switching off the

pathways involved in cancer cell survival using chemicals before irradiation, while sparing normal cells will be an effective way to protect the normal cells. In contrast to normal cells, cancer cells mostly fail to activate damage sensor proteins involved in DNA repair pathways are often dysfunctional. DNA repair deficiency in cancer cells stimulates mutagenesis and further leads to tumorigenesis. However, at the same time these tumor cells are prone to the DNA damage by chemotherapy or radiotherapy treatments. From a clinical perspective, a good chemical radioprotectors must target this DNA repair pathways to kill the cancer cells differentially while protecting the normal cells. Therefore, a potential radio-protective agent should have more profound differential radiosensitizing effect on cancer cells include cell cycle arrest, apoptosis, direct and indirect effects on DNA bases, repair proteins and tumor vasculature. Understanding this problem at a molecular level, while reducing any unwanted deposition of radiation doses to the surrounding normal cells/tissues is a central pursuit in radiobiological research.

Repopulation of cells in critical normal tissues between individual dose fractions of either radiotherapy or chemotherapy is an important factor for the recovery or retention of normal organ function, thereby improving tolerance to treatment. Normal cells have a tremendous ability to repair DNA against radiation damage by activating "damage sensor" proteins such as activating ATM, and ATR, checkpoint kinase 1 and 2 (CHK1 and CHK2) or p53 [52-54]. PI3K also plays a role in the integral functions for noncancerous (normal) cells repopulation [55] along with the DNA repair proteins. Once DNA damage detected and sensor proteins activated, each lesion can be repaired by at least one of the six major DNA-repair pathways: BER (base excision repair); NER (nucleotide excision repair); DR (direct repair); MMR (mismatch repair); HR (homologous recombination) or NHEJ (non-homologous end joining) pathways [56, 57], if not, cell death occurs due to residual or misrepaired DNA double-strand breaks [58]. In contrast to normal cells, in cancer cells DNA-repair path-ways are dysfunctional and this may make tumor cells prone to the DNA-damaging agents such as radiotherapy and/or chemotherapy [53, 59, 60]. Furthermore understanding the biological mechanisms involved in signaling pathway(s) for resistance, cell cycle check-

points, DNA damage and repair, anti-angiogenesis could increase the therapeutic response of tumor microenvironment while sparing surrounding normal tissues. However, work on radioprotective chemicals started several decades ago in the USA, at the inception of the Manhattan Project and the available literature on the topic is enormous, this review focuses only on those relevant and potential agents which are of clinical importance with their mechanism of radioprotection.

In recent years, strategies to improve radiotherapy therefore aim to increase the effect on tumor while limiting the damage to the adjacent normal cells without sensitizing the normal tissues and ultimately without protecting the tumors to the radiation treatment. Despite the availability of various sophisticated technical improvements in radiotherapy [61, 62], treatment planning techniques and stereotactic body radiation therapy (SBRT) modalities, normal tissue toxicity remains a dose-limiting problem in therapeutic programs [19, 63]. The development of radioprotectors or radioprotectants that protect normal tissues surrounding the tumor cells against radiation damage is currently the subject of intense research [64]. Radioprotectors are chemicals and most of the radioprotective compounds are antioxidants, can be given before or at the time of radiation treatment of cancer control. In general, radioprotectors are drugs that are designed with the intent of minimizing the risk of clonogenic death of normal (noncancerous) cells from the damage caused by radiation. These agents also promote the repair of normal cells that are exposed to radiation.

In recent years many chemical agents that have been postulated which protects the normal cells by targeting newly identified molecular and physiological pathways. Therefore, detailed understanding of the pathways influenced by the radioprotectors, as well as identification and characterization of the participating proteins will significantly advance our ability to unravel the complex processes leading to the development of new drugs that protect normal cells/tissues. Despite technical improvements, no radioprotective drug available today shows all the requisite qualities to be an ideal radioprotector and many patients still suffer from recurrent disease after radiotherapy. An ideal radioprotector is relatively non-toxic to normal cells and easy to

administer without compromising the therapeutic effects of radiation treatment for cancer patients. For years, many radioprotective compounds have been developed, a majority of them designed to reduce the levels of radiation-induced free radicals within the cells. Indeed, after several decades of preclinical and clinical research, the first and only approved radioprotective drug by U.S. Food and Drug Administration (FDA) is amifostine, being used in clinical practice.

Amifostine is a prodrug (inactive form) which belongs to a general class of cytoprotective (cell-protecting) agents. In the body, amifostine is converted into an active thiol metabolite WR1065, which helps to protect the cells from DNA damage by scavenging free radicals [65]. The conversion of amifostine to WR1065 is catalyzed by alkaline phosphatase which is a pH dependent and occurring more rapidly in alkaline pH, tumor cells are acidic in nature [66, 67]. Once dephosphorylated, it can freely diffuse mostly into normal cells and can act as a free radical scavenger. Furthermore, the lower concentration of membrane-bound alkaline phosphatase and lower pH (acidic) in tumor microenvironments contributes to a relatively low concentration of active chemoprotectant in malignant tissues and therefore providing a selective cytoprotection of normal tissues [68, 69] around the irradiation field. Since tumors are relatively in hypoxic condition due to the poor vasculature, thus resulting in comparative hypoxia and a low (acidic) interstitial pH than normal tissues [70-73]. Therefore normal tissues can absorb greater level of amifostine than the tumor tissues, and furthermore cancer cells are not protected. The half-life of amifostine is approximately 9 minutes, whereas that of the active metabolite, WR1065, is approximately 15 minutes [74]. Thus, the schedule of amifostine administration is potentially an important factor for optimum efficacy for the radiation treatment to the cancer patients.

In both normal and cancer cells, ionizing radiation has been shown to generate reactive oxygen species (ROS), mitochondrial respiratory chain is a major source of reactive oxygen species under various pathological conditions [75-77]. Furthermore, mutation in mitochondrial DNA (mtDNA) increased the metastatic potential of tumor cells [78]. ROS cause oxidative damage to DNA, proteins, lipids, and other cellular

components and therefore pose a significant cellular stress [79] and recent studies suggest that this biochemical property of cancer cells can be exploited for therapeutic benefits. Therefore targeting mitochondria is one of an essential step to therapeutic approach for the cancer control [80]. However, in cancer cells aberrant metabolism and protein translation generate abnormally high levels of ROS [81]. Agents that enhance ROS production, therefore, are expected to cause further stress overload in cancer cells by increase in DNA damage. Since ROS act as a mediator of the cellular damage induced by radiation, compounds that involved in the regulation of ROS may be of great interest in the protection of normal cells against radiation induced damage. For e.g. it has been shown that dichloroacetate, which inhibits pyruvate dehydrogenase kinase (PDK) and therefore stimulates mitochondrial oxidative phosphorylation and ROS production, such mechanisms can selectively increase DNA damage related apoptosis in cancer cells but not in normal cells [82]. Similarly, reducing the cellular ROS buffering capacity through the inhibition of glutamate-cysteine ligase (a rate-limiting enzyme in cellular glutathione synthesis) can markedly increase the radiosensitivity of cancer cells [83].

In humans, as in most animal cells, maintenance and expression of mtDNA are essential for the normal cell survival and metabolism. Manganese superoxide dismutase (MnSOD) exists exclusively in the mitochondria and scavenges toxic superoxide radicals produced by radiation. Therefore it has been proposed that MnSOD play a role in protecting cells against ROS damage during the radiation exposure. Lee et al. [84] showed mitochondrial damage such as altered permeability transition, increase in accumulation of reactive oxygen species, reduction of ATP production and morphological change induced by radiation in cells and mice were protected by the treatment of MnSOD. Preclinical studies in mouse have demonstrated that the expression of human MnSOD transgene confers protection of normal tissues from ionizing irradiation damage and also radiosensitizing the tumor cells [85-87]. Administration of manganese superoxide dismutase plasmid liposomes (MnSOD-PL) carrying DNA damage control genes has been demonstrated to provide local radiation protection to the lung, esophagus, oral cavity, urinary bladder and intestine [88, 89]. Therefore, over expression of MnSOD

has been shown to sensitize the cancer cells to radiation, while differentially over expression in normal cells protect from irradiation [90].

In cancer patients undergoing radiation treatment, highly proliferating organs like intestine and bone marrow limits its beneficial effects. Using mouse model, Burdelya and colleagues recently tested a new approach to protect the intestine from irradiation-induced injury. Burdelya et al. [91] developed an NF-kB-activating CBLB502 a polypeptide drug by modifying a small fragment of a *Salmonella flagella*. CBLB502 is a bioengineered derivative of a microbial protein derived from flagellin. Human immune response to flagellin can be explained by two facts: 1. flagellin is an extremely abundant protein in flagellated bacteria, 2. a specific innate immune receptor that recognizes flagellin is toll like receptor-5 (TLR-5). Most TLRs activates NF-κB, which transcri-ptionally regulates a diverse array of genes. These include cytokines, chemokines and their respective receptors, which are associated with the important role of NF-κB in the inflammatory response, together with genes regulating cell survival, proliferation, cell adhesion in the cellular microenviroment [32, 92-94]. All these events activated by TLR-NF-kB protect normal cells by preventing death of the cell. Although the NF-κB gene targets may be similar between normal and cancer cells, the difference is the 'appropriateness' of the signal and their regulation [95]. For example, in tumor cell NF-κB targets may show sustained induction of their expression, resulting from the loss of negative feedback control mechanisms. Furthermore, NF-κB-transcription dependent gene expressions can either promote growth and survival of cancer cells or contribute towards tumor suppressor mechanisms, depending on the status of tumor cell for e.g loss of key tumor suppressors such as p53 or PTEN can drive NF-κB towards oncogenic and tumor-promoting activity [96]. Therefore, NF-κB pathway can be exploited as a target for normal cell survival from radiation injury.

Burdelya et al. [91] injected CBLB502 into mice before total body irradiation. The treatment prevented radiation induced mortality or completely protected the animals against radiation induced damage. Another interesting application of the drug is that it protects normal cells without diminishing the therapeutic antitumor effect of radiation and further without promoting radia-

tion induced carcinogenicity in the tumor cell injected mice. CBLB502 uniquely showed combined properties of supportive care (radiotherapy adjuvant) and anticancer agent, both mediated via the activation of TLR-5 signaling in normal tissues or in tumor, respectively [96]. Thus, TLR-5 agonists CBLB502 could potentially protect normal cells, while improving the therapeutic index of radiotherapy when large and actively proliferating organs like intestine or bone marrow are considered as a dose limiting factors. MnSOD gene therapy and CBLB502 are still in the early stages of development but in the available radioprotectors it could allow for safer and more effective radiation treatment while protecting the normal cells.

Conclusion

With the improved clinical outcomes of cancer treatment, minimizing radiotherapy related toxicities on normal cells/tissues microenvironment is a priority. Although the overall cancer treatment time allows the repopulation of cells in normal tissues, repopulation of surviving cancer cells also occurs, thereby increasing the number of cancer cells that must be eradicated with the minimal damage to the normal cells. In recent years, development of radioprotective agents that spare healthy tissues during radiotherapy and also to protect against accidental, occupational and terrorist-initiated radiation exposure is the subject of intense research. Though in radiotherapy accurate delivery of radiation dose has been greatly improved over the past 2-3 decades, a major aim in the field of radiotherapy is to identify efficient ways of killing the cancer cells to improve the cancer cure rates, while decreasing the toxicities of normal cells is the greatest challenge in the field of radiation oncology of modern medicine. On the other hand, radiation injuries to the normal cells/tissues represent a significant challenge, because pathophysiology of radiation interactions on cellular communications between cancer and normal cells are not completely understood as yet. The increased nature of cancer cell proliferation with the decrease in cell division time is highly suggestive that the rapid cell proliferation might be sensitive to radiation induced cell killing effect. However, over the past decade, molecular biological approaches applied to radiobiological questions have uncovered several mechanisms by which cancer and normal cells differently respond to ionizing ra-

diation. In recent years, a number of potential molecular targets has been identified and the hope is that translating these basic research findings to enhancing the toxicity of ionizing radiation on the tumor cells for therapeutic exploitation will be able to target only cancer cells with the goal of sharply reducing the number of people suffering and dying from cancer disease.

Address correspondence to: Dr. Baskar R, Department of Radiation Oncology, Molecular Radiation Biology Laboratory, Division of Cellular and Molecular Research, National Cancer Centre, 11- Hospital Drive, Singapore-169610, Republic of Singapore Tel: +65-6436 8315; Fax: +65-6222 8675; E-mail: r.baskar@nccs.com.sg

References

[1] Jemal A, Center MM, DeSantis C, Ward EM. Global patterns of cancer incidence and mortality rates and trends. Cancer Epidemiol Biomarkers Prev 2010; 19: 1893-1907.

[2] Thun MJ, DeLancey JO, Center MM, Jemal A, Ward EM. The global burden of cancer: priorities for prevention. Carcinogenesis 2010; 31: 100-110.

[3] Weckermann D, Müller P, Wawroschek F, Harzmann R, Riethmüller G, Schlimok G. Disseminated cytokeratin positive tumor cells in the bone marrow of patients with prostate cancer: detection and prognostic value. J Urol 2001; 166: 699-703.

[4] Karrison TG, Ferguson DJ, Meier P. Dormancy of mammary carcinoma after mastectomy. J Natl Cancer Inst 1999; 91: 80-85.

[5] Pfitzenmaier J, Ellis WJ, Arfman EW, Hawley S, McLaughlin PO, Lange PH, Vessella RL. Telomerase activity in disseminated prostate cancer cells. BJU Int 2006; 97: 1309-1313.

[6] Aguirre-Ghiso JA. Models mechanisms and clinical evidence for cancer dormancy. Nat Rev Cancer 2007; 7: 834-846.

[7] International Agency for Research on Cancer (IARC): GLOBOCAN 2008, Cancer incidence and mortality worldwide. Lyon, France, 2010.

[8] Hamilton AS, Mack TM. Puberty and genetic susceptibility to breast cancer in a case-control study in twins. N Engl J Med 2003; 348: 2313-2322.

[9] Anand P, Kunnumakkara AB, Sundaram C, Harikumar KB, Tharakan ST, Lai OS, Sung B, Aggarwal BB. Cancer is a preventable disease that requires major lifestyle changes. Pharm

Res 2008; 25: 2097-2116.

[10] Bernier J, Hall EJ, Giaccia A. Radiation oncology: a century of achievements. Nat Rev Cancer 2004; 4: 737-747.

[11] Bentzen SM. Preventing or reducing late side effects of radiation therapy: radiobiology meets molecular pathology. Nat Rev Cancer 2006; 6: 702-713.

[12] Das P, Cantor SB, Parker CL, Zampieri JB, Baschnagel A, Eng C, Delclos ME, Krishnan S, Janjan NA, Crane CH. Long-term quality of life after radiotherapy for the treatment of anal cancer. Cancer 2010; 116: 822-829.

[13] Ringborg U, Bergqvist D, Brorsson B, Cavallin-Ståhl E, Ceberg J, Einhorn N, Frödin JE, Järhult J, Lamnevik G, Lindholm C, Littbrand B, Norlund A, Nylén U, Rosén M, Svensson H, Möller TR. The Swedish Council on Technology Assessment in Health Care: systematic overview of radiotherapy for cancer including a prospective survey of radiotherapy practice in Sweden 2001-summary and conclusions. Acta Oncol 2003; 42: 357-365.

[14] Hodges PC. The life and times of E´mil H. Grubbe´. Chicago: University of Chicago Press 1964; pp: 135.

[15] Connell PP, Hellman S. Advances in radiotherapy and implications for the next century: a historical perspective. Cancer Res 2009; 69: 383-392.

[16] Howlader N, Noone AM, Krapcho M, Neyman N, Aminou R, Waldron W, Altekruse SF, Kosary CL, Ruhl J, Tatalovich Z, Cho H, Mariotto A, Eisner MP, Lewis DR, Chen HS, Feuer EJ, Cronin KA, Edwards BK (eds). SEER Cancer Statistics Review, 1975-2008, National Cancer Institute. Bethesda, MD, http://seer.cancer.gov/csr/1975_2008/, based on November 2010 SEER data submission, posted to the SEER web site, 2011.

[17] Travis LB, Ng AK, Allan JM, Pui CH, Kennedy AR, Xu XG, Purdy JA, Applegate K, Yahalom J, Constine LS, Gilbert ES, Boice JD Jr. Second malignant neoplasms and cardiovascular disease following radiotherapy. J Natl Cancer Inst 2012; 104: 357-370.

[18] Barcellos-Hoff MH, Park C, Wright EG. Radiation and the microenvironment - tumorigenesis and therapy. Nat Rev Cancer 2005; 5: 867-875.

[19] Baskar R. Emerging role of radiation induced bystander effects: cell communications and carcinogenesis. Genome Integr 2010; 1: 13.

[20] Stone HB, Coleman CN, Anscher MS, McBride WH. Effects of radiation on normal tissue: consequences and mechanisms. Lancet Oncol 2003; 4: 529-536.

[21] Maxhimer JB, Soto-Pantoja DR, Ridnour LA, Shih HB, Degraff WG, Tsokos M, Wink DA, Isenberg JS, Roberts DD. Radioprotection in normal tissue and delayed tumor growth by blockade of CD47 signaling. Sci Transl Med 2009; 1: 3ra7.

[22] Anscher MS, Thrasher B, Rabbani Z, Teicher B, Vujaskovic Z. Antitransforming growth factor-beta antibody 1D11 ameliorates normal tissue damage caused by high-dose radiation. Int J Radiat Oncol Biol Phys 2006; 65: 876-881.

[23] Shuryak I, Hahnfeldt P, Hlatky L, Sachs RK, Brenner DJ. A new view of radiation-induced cancer: integrating short- and long-term processes. Part II: second cancer risk estimation. Radiat Environ Biophys 2009; 48: 275-286.

[24] Hunger SP, Lu X, Devidas M, Camitta BM, Gaynon PS, Winick NJ, Reaman GH, Carroll WL. Improved survival for children and adolescents with acute lymphoblastic leukemia between 1990 and 2005: a report from the children's oncology group. J Clin Oncol 2012, Mar 12 [Epub ahead of print].

[25] Tuan JK, Ha TC, Ong WS, Siow TR, Tham IW, Yap SP, Tan TW, Chua ET, Fong KW, Wee JT. Late toxicities after conventional radiation therapy alone for nasopharyngeal carcinoma. Radiother Oncol 2012; 30: 1663-1669.

[26] Sachs RK, Brenner DJ. Solid tumor risks after high doses of ionizing radiation. Proc Natl Acad Sci USA 2005; 102: 13040-13045.

[27] Hall EJ. Intensity-modulated radiation therapy, protons, and the risk of second cancers. Int J Radiat Oncol Biol Phys 2006; 65: 1-7.

[28] Armstrong GT, Liu W, Leisenring W, Yasui Y, Hammond S, Bhatia S, Neglia JP, Stovall M, Srivastava D, Robison LL. Occurrence of multiple subsequent neoplasms in long-term survivors of childhood cancer: a report from the childhood cancer survivor study. J Clin Oncol 2011; 29: 3056-3064.

[29] Ng J, Shuryak I, Xu Y, Clifford Chao KS, Brenner DJ, Burri RJ. Predicting the risk of secondary lung malignancies associated with whole-breast radiation therapy. Int J Radiat Oncol Biol Phys 2012 Jan 13 [Epub ahead of print].

[30] Spitz DR, Azzam EI, Li JJ, Gius D. Metabolic oxidation/reduction reactions and cellular responses to ionizing radiation: a unifying concept in stress response biology. Cancer Metastasis Rev 2004; 23: 311-322.

[31] Cann KL, Hicks GG. Regulation of the cellular DNA double-strand break response. Biochem Cell Biol 2007; 85: 663-674.

[32] Kim HJ, Hawke N, Baldwin AS. NF-κB and IKK as therapeutic targets in cancer. Cell Death Differ 2006; 13: 738-747.

[33] Dorr W. Three A's of repopulation during fractionated irradiation of squamous epithelia: asymmetry loss, acceleration of stem-cell divisions and abortive divisions. Int J Radiat Biol 1997; 72: 635-643.

[34] Iliakis G, Wang Y, Guan J, Wang H. DNA damage checkpoint control in cells exposed to ionizing radiation. Oncogene 2003; 22: 5834-5847.

[35] Ward JF. The complexity of DNA damage: relevance to biological consequences. Int J Radiat Biol 1995; 66: 427-432.

[36] Radford IR. The level of induced DNA double-strand breakage correlates with cell killing after X-irradiation. Int J Radiat Biol Relat Stud Phys Chem Med 1985; 48: 45-54.

[37] Jorgensen TJ. Enhancing radiosensitivity: targeting the DNA repair pathways. Cancer Biol Ther 2009; 8: 665-670.

[38] Engelman JA. Targeting PI3K signalling in cancer: opportunities, challenges and limitations. Nat Rev Cancer 2009; 9: 550-562.

[39] Castellano E, Downward J. RAS interaction with PI3K: more than just another effector pathway. Genes and Cancer 2011; 2: 261-274.

[40] Schuurbiers OC, Kaanders JH, van der Heijden HF, Dekhuijzen RP, Oyen WJ, Bussink J. The PI3-K/AKT-pathway and radiation resistance mechanisms in non-small cell lung cancer. J Thorac Oncol 2009; 4: 761-767.

[41] Jain RK. Normalization of tumor vasculature: an emerging concept in antiangio-genic therapy. Science 2005; 307: 58-62.

[42] Pries AR, Cornelissen AJ, Sloot AA, Hinkeldey M, Dreher MR, Höpfner M, Dewhirst MW, Secomb TW. Structural adaptation and heterogeneity of normal and tumor microvascular networks. PLoS Comput Biol 2009; 5: e1000394.

[43] Dewhirst MW, Cao Y, Moeller B. Cycling hypoxia and free radicals regulate angiogenesis and radiotherapy response. Nat Rev Cancer 2008; 8: 425-437.

[44] Semenza GL. Hypoxia-inducible factors in physiology and medicine. Cell 2012; 148: 399-408.

[45] Semenza GL. Targeting HIF-1 for cancer therapy. Nat Rev Cancer 2003; 10: 721-732.

[46] Moeller BJ, Richardson RA, Dewhirst MW. Hypoxia and radiotherapy: opportunities for improved outcomes in cancer treatment. Cancer Metastasis Rev 2007; 26: 241-248.

[47] Hanahan D, Weinberg RA. The hallmarks of cancer. Cell 2000; 100: 57-70.

[48] Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell 2011; 144: 646-674.

[49] Weinstein IB. Cancer. Addiction to oncogenes-the achilles heal of cancer. Science 2002; 297: 63-64.

[50] Weinstein IB, Joe AK. Mechanisms of disease: oncogene addiction-a rationale for molecular targeting in cancer therapy. Nat Clin Pract Oncol 2006; 3: 448-457.

[51] Sharma SV, Settleman J. Oncogene addiction: setting the stage for molecularly targeted cancer therapy. Genes Dev 2007; 21: 3214-3231.

[52] Ch'ang HJ, Maj JG, Paris F, Xing HR, Zhang J, Truman JP, Cardon-Cardo C, Haimovitz-Friedman A, Kolesnick R, Fuks Z. ATM regulates target switching to escalating doses of radiation in the intestines. Nat Med 2005; 11: 484-490.

[53] Helleday T, Petermann E, Lundin C, Hodgson B, Sharma RA. DNA repair pathways as targets for cancer therapy. Nat Rev Cancer 2008; 8: 193-204.

[54] Helleday T. DNA repair as treatment target. Eur J Cancer 2011; 47: S333-335.

[55] Courtney KD, Corcoran RB, Engelman JA. The PI3K pathway as drug target in human cancer. J Clin Oncol 2010; 28: 1075-1083.

[56] Jalal S, Earley JN, Turchi JJ. DNA repair: from genome maintenance to biomarker and therapeutic target. Clin Cancer Res 2011; 17: 6973-6984.

[57] Neher TM, Turchi JJ. Current advances in DNA repair: regulation of enzymes and pathways involved in maintaining genomic stability. Antioxidants Redox Signaling 2011; 14: 2461-2464.

[58] Bedford JS, Mitchell JB, Griggs HG, Bender MA. Radiation-induced cellular reproductive death and chromosome aberrations. Radiat Res 1978; 76: 573-586.

[59] Lord CJ, Ashworth A. The DNA damage response and cancer therapy. Nature 2012; 481: 287-294.

[60] Postel-Vinay S, Vanhecke E, Olaussen KA, Lord CJ, Ashworth A, Soria JC. The potential of exploiting DNA-repair defects for optimizing lung cancer treatment. Nat Rev Clin Oncol 2012; 9: 144-155.

[61] Jaffray DA. Emergent technologies for 3-dimensional image-guided radiation delivery. Semin Radiat Oncol 2005; 15: 208-216.

[62] Baskar R, Lee KA, Yeo R, Yeoh KW. Cancer and radiation therapy: current advances and future directions. Int J Med Sci 2012; 9: 193-199.

[63] Bhide SA, Nutting CM. Recent advances in radiotherapy. BMC Med 2010; 8: 25.

[64] Mettler Jr FA, Brenner D, Coleman CN, Kaminski JM, Kennedy AR, Wagner LK. Can radiation risks to patients be reduced without reducing radiation exposure? The status of chemical radioprotectants. Am J Roentgenol 2011; 196: 616-618.

[65] Koukourakis MI. Amifostine in clinical oncology: current use and future applications. Anticancer Drugs 2002; 13: 181-209.

[66] Gatenby R A, Gawlinski ET, Gmitro AF, Kaylor B, Gillies RJ. Acid-mediated tumor invasion: a multidisciplinary study. Cancer Res 2006; 66: 5216-5223.

[67] Webb BA, Chimenti M, Jacobson MP, Barber DL. Dysregulated pH: a perfect storm for cancer progression. Nat Rev Cancer 2011; 11: 671-677.

[68] Spencer CM and Goa KL. Amifostine: a review of its pharmacodynamic and pharmacokinetics properties, and therapeutic potential as a radioprotector and cytotoxic chemoprotector. Drugs 1995; 50: 1001-1031.

[69] Kemp G, Rose P, Lurain J, Berman M, Manetta A, Roullet B, Homesley H, Belpomme D, Glick J. Amifostine pretreatment for protection against cyclophosphamide-induced and cisplatin-induced toxicities: results of a randomized control trial in patients with advanced ovarian can-cer. J Clin Oncol 1996; 14: 2101-2112.

[70] Tannock IF, Rotin D. Acid pH in tumors and its potential for therapeutic exploitation. Cancer Res 1989; 49: 4373-4384.

[71] Vaupel P. Hypoxia and aggressive tumor phenotype: implications for therapy and prognosis. The Oncologist 2008; 13: 21-26.

[72] Wilson WR, Hay MP. Targeting hypoxia in cancer therapy. Nat Rev Cancer 2011; 11: 393-400.

[73] Webb BA, Chimenti M, Jacobson MP, Barber DL. Dysregulated pH: a perfect storm for cancer progression. Nat Rev Cancer 2011; 11: 671-677.

[74] McKibbin T, Panetta JC, Fouladi M, Gajjar A, Bai F, Okcu MF, Stewart CF. Clinical pharmacokinetics of amifostine and WR1065 in pediatric patients with medulloblastoma. Clin Cancer Res 2010; 16: 1049-1057.

[75] Chen Q, Vazquez EJ, Moghaddas S, Hoppel CL, Lesnefsky EJ. Production of reactive oxygen species by mitochondria: central role of complex III. J Biol Chem 2003; 278: 36027-36031.

[76] Balaban RS, Nemoto S, Finkel T. Mitochondria, oxidants, and aging. Cell 2005; 120: 483-495.

[77] Murphy MP. How mitochondria produce reactive oxygen species. Biochem J 2009; 417: 1-13.

[78] shikawa K, Takenaga K, Akimoto M, Koshikawa N, Yamaguchi A, Imanishi H, Nakada K, Honma Y, Hayashi J. ROS-generating mitochondrial DNA mutations can regulate tumor cell metastasis. Science 2008; 320: 661-664.

[79] Avery SV. Molecular targets of oxidative stress. Biochem J 2011; 434: 201-210.

[80] Fulda S, Galluzzi L, Kroemer G. Targeting mitochondria for cancer therapy. Nat Rev Drug Discov 2010; 9: 447-464.

[81] Cairns RA, Harris IS, Mak TW. Regulation of cancer cell metabolism. Nat Rev Cancer 2011; 11: 85-95.

[82] Bonnet S, Archer SL, Allalunis-Turner J, Haromy A, Beaulieu C, Thompson R, Lee CT, Lopaschuk GD, Puttagunta L, Bonnet S, Harry G, Hashimoto K, Porter CJ, Andrade MA, Thebaud B, Michelakis ED. A mitochondria-K+ channel axis is suppressed in cancer and its normalization promotes apoptosis and inhibits cancer growth. Cancer Cell 2007; 11: 37-51.

[83] Diehn M, Cho RW, Lobo NA, Kalisky T, Dorie MJ, Kulp AN, Qian D, Lam JS, Ailles LE, Wong M, Joshua B, Kaplan MJ, Wapnir I, Dirbas FM, Somlo G, Garberoglio C, Paz B, Shen J, Lau SK, Quake SR, Brown JM, Weissman IL, Clarke MF. Association of reactive oxygen species levels and radioresistance in cancer stem cells. Nature 2009; 458: 780-783.

[84] Lee YJ, Jeong SY, Karbowski M, Smith C L, Youle RJ. Roles of the mammalian mitochondrial fission and fusion mediators Fis1, Drp1, and Opa1 in apoptosis. Mol Biol Cell 2004; 15: 5001-5011.

[85] Borrelli A, Schiattarella A, Mancini R, Morrica B, Cerciello V, Mormile M, d'Alesio V, Bottalico L, Morelli F, D'Armiento M, D'Armiento FP, Mancini A. A recombinant MnSOD is radioprotective for normal cells and radiosensitizing for tumor cells. Free Radic Biol Med 2009; 46: 110-116

[86] Epperly MW, Wang H, Jones JA, Dixon T, Montesinos CA, Greenberger JS. Antioxidant-chemoprevention diet ameliorates late effects of total-body irradiation and supplements radio-protection by MnSOD-plasmid liposome administration. Radiat Res 2011; 175: 759-765.

[87] Tarhini AA, Belani CP, Luketich JD, Argiris A, Ramalingam SS, Gooding W, Pennathur A, Petro D, Kane K, Liggitt D, Championsmith T, Zhang X, Epperly MW, Greenberger JS. A phase I study of concurrent chemotherapy (paclitaxel and carboplatin) and thoracic radiotherapy with swallowed manganese superoxide dismutase plasmid liposome protection in patients with locally advanced stage III non-small-cell lung cancer. Hum Gene Ther 2011; 22: 336-342.

[88] Greenberger JS, Epperly MW. Antioxidant gene therapeutic approaches to normal tissue radio-protection and tumor radiosensitization. In Vivo 2007; 21: 141-146.

[89] Niu Y, Wang H, Wiktor-Brown D, Rugo R, Shen H, Huq MS, Engelward B, Epperly M, Greenberger JS. Irradiated esophageal cells are protected from radiation-induced recombination by MnSOD gene therapy. Radiat Res 2010; 173: 453-461

[90] Borrelli A, Schiattarella A, Mancini R, Morrica B, Cerciello V, Mormile M, d'Alesio V, Bottalico L, Morelli F, D'Armiento M, D'Armiento FP, Mancini A. A recombinant MnSOD is radioprotective for normal cells and radiosensitizing for tumor cells. Free Radic Biol Med 2009; 46: 110-116.

[91] Burdelya LG, Krivokrysenko VI, Tallant TC, Strom E, Gleiberman AS, Gupta D, Kurnasov OV, Fort FL, Osterman AL, Didonato JA, Feinstein E, Gudkov AV. An agonist of toll-like receptor 5 has radioprotective activity in mouse and primate models. Science 2008; 320: 226-230.

[92] Kumar A, Takada Y, Boriek AM, Aggarwal BB. Nuclear factor-κB: its role in health and disease. J Mol Med 2004; 82: 434-448.

[93] Fan Y, Dutta J, Gupta N, Fan G, Gelinas C. Regulation of programmed cell death by NF-κB and its role in tumorigenesis and therapy. Adv Exp Med Biol 2008; 615: 223-250.

[94] Perkins ND, Gilmore TD. Good cop, bad cop: the different faces of NF-κB. Cell Death Differ 2006; 13: 759-772.

[95] Perkins ND. The diverse and complex roles of NF-κB subunits in cancer. Nat Rev Cancer 2012; 12: 121-132.

[96] Burdelya LG, Gleiberman AS, Toshkov I, Aygun-Sunar S, Bapardekar M, Manderscheid-Kern P, Bellnier D, Krivokrysenko VI, Feinstein E, Gudkov AV. Toll-like Receptor 5 Agonist Protects Mice from Dermatitis and Oral Mucositis Caused by Local Radiation: Implications for Head-and-Neck Cancer Radiotherapy. Int J Radiat Oncol Biol Phys 2012; 83: 228-234.