

Radioprotectors and Mitigators of Radiation-Induced Normal Tissue Injury

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ABSTRACT

Radiation is used in the treatment of a broad range of malignancies. Exposure of normal tissue to radiation may result in both acute and chronic toxicities that can result in an inability to deliver the intended therapy, a range of symptoms, and a decrease in quality of life. Radioprotectors are compounds that are designed to reduce the damage in normal tissues caused by radiation. These compounds are often antioxidants and must be present be-

fore or at the time of radiation for effectiveness. Other agents, termed mitigators, may be used to minimize toxicity even after radiation has been delivered. Herein, we review agents in clinical use or in development as radioprotectors and mitigators of radiation-induced normal tissue injury. Few agents are approved for clinical use, but many new compounds show promising results in preclinical testing. *The Oncologist* 2010;15:360–371

INTRODUCTION

Radiotherapy is commonly used as a component of therapy for a wide range of malignant conditions. It is estimated that half of all cancer patients will receive radiotherapy during the course of their treatment for cancer [1]. Radiotherapy is frequently used to achieve local or regional control of malignancies either alone or in combination with other modalities such as chemotherapy or surgery.

Irradiation of noncancerous “normal” tissues during the course of therapeutic radiation can result in a range of side

effects including self-limited acute toxicities, mild chronic symptoms, or severe organ dysfunction. The likelihood of developing these complications relates to the volume of an organ irradiated, the radiation dose delivered, fractionation of the delivered dose, the delivery of radiation modifiers, and individual radiosensitivity. Efforts to reduce the toxicities associated with therapeutic radiation have centered on both technological improvements in radiation delivery and chemical modifiers of radiation injury.

The use of technology to reduce normal tissue toxicity

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includes techniques such as conformal radiotherapy, intensity-modulated radiotherapy, image-guided radiotherapy, and proton radiotherapy. Each of these aims to reduce the volume of normal tissue exposed to high doses of radiation compared with conventional therapies, thus reducing the risk for normal tissue toxicity. Studies of these modalities have shown that better normal tissue dose distributions and side effect profiles can be obtained using these technologies. Although improvements have been realized in this regard, normal tissue toxicity remains a limiting factor in the treatment of many diseases with radiation therapy. Based on the intimate relationship between tumors and their normal host tissues and surrounding critical structures and the need to irradiate clinically uninvolved normal tissue margins that are potentially contaminated with microscopic disease, it is anticipated that normal tissue toxicity will remain a concern for therapeutic radiation.

An alternative mechanism to reduce normal tissue toxicity is the use of radiation modifiers/protectors, agents that when present prior to or shortly after radiation exposure alter the response of normal tissues to irradiation. This approach has also been viewed as an attractive countermeasure for possible nuclear/radiological terrorism. To be useful in the radiotherapy clinic, radioprotectors should ideally have several characteristics that relate to the ability of the agent to improve the therapeutic ratio. First, the agent should be selective in protecting normal tissues from radiotherapy without protecting tumor tissue, otherwise no benefit in the therapeutic index will be realized. Second, the agent should be delivered with relative ease and with minimal toxicity. Finally, the agent should protect normal tissues that are considered sensitive such that acute or late toxicities in these tissues are either dose-limiting or responsible for a significant reduction in quality of life (i.e., mucositis, pneumonitis, myelopathy, xerostomia, proctitis, and leukencephalopathy). Although a number of compounds have been described that meet most or all of these criteria in preclinical studies or in early clinical trials, only amifostine is currently in clinical use. Herein, we provide a classification of agents that are being evaluated to prevent or treat normal tissue injury, describe the events that occur after radiation that are responsible for the injury to normal tissue, and discuss some agents in development as radiation protectors and radiation mitigators.

CLASSIFICATION OF AGENTS

In general, chemical/biological agents used to alter normal tissue toxicity from radiation can be broadly divided into three categories based on the timing of delivery in relation to radiation: chemical radioprotectors, mitigators, and treatment [2]. Agents delivered prior to or at the time of irradi-

ation with the intent of preventing or reducing damage to normal tissues are termed radioprotectors. Agents delivered at the time of irradiation or after irradiation is complete, but prior to the manifestation of normal tissue toxicity, are described as mitigators of normal tissue injury. Finally, agents delivered to ameliorate established normal tissue toxicity are considered treatments. There is a growing body of literature describing radioprotection or mitigation with a variety of agents after total body exposures or localized exposures. A complete and exhaustive review of these agents is outside the scope of this review. Herein, we briefly highlight several classes of agents that have been described as radiation protectors and mitigators, and attempt to focus on agents with demonstrated or anticipated clinical usefulness for therapeutic radiation exposures. Treatment of established radiation normal tissue injury is outside the scope of this work.

An understanding of the events occurring during and shortly after irradiation of tissues and cells is important to understanding the mechanism of action of radioprotectors and mitigators. Figure 1 shows the sequence of events in cells and tissues following radiation exposure. Radiation can damage cells/tissues by both direct and indirect actions [3]. The term “direct effects” describes radiation directly causing irreparable damage to critical targets within the cell, such as DNA. The term “indirect effects” describes the situation in which radiation interacts with other molecules of the cell that are not critical targets but are close enough to pass on this damage, typically in the form of free radicals. Because mammals are composed of roughly 80% water, indirect effects involve the production of radiolysis products of water, in particular, the hydroxyl free radical, which is a potent oxidant capable of breaking chemical bonds, initiating lipid peroxidation, in the nano- to microsecond time-frame [4].

After DNA damage has occurred, a number of processes occur in the damaged cell, tissue, or organism (Fig. 1), including activation of DNA repair, activation of signal transduction, expression of radiation response genes, stimulation of proliferation, and initiation and perpetuation of inflammation. These pathways can be important for cell or tissue recovery after radiation exposure but may also play a role in the development of toxicity. Mitigators of radiation injury may target these pathways to prevent or reduce the expression of toxicity.

Radioprotectors: Reducing Agents/Free Radical Scavengers

As described above, free radicals are responsible for perpetuating a large amount of the damage caused by ionizing radiation. Therefore, for an agent to protect cells from pri-

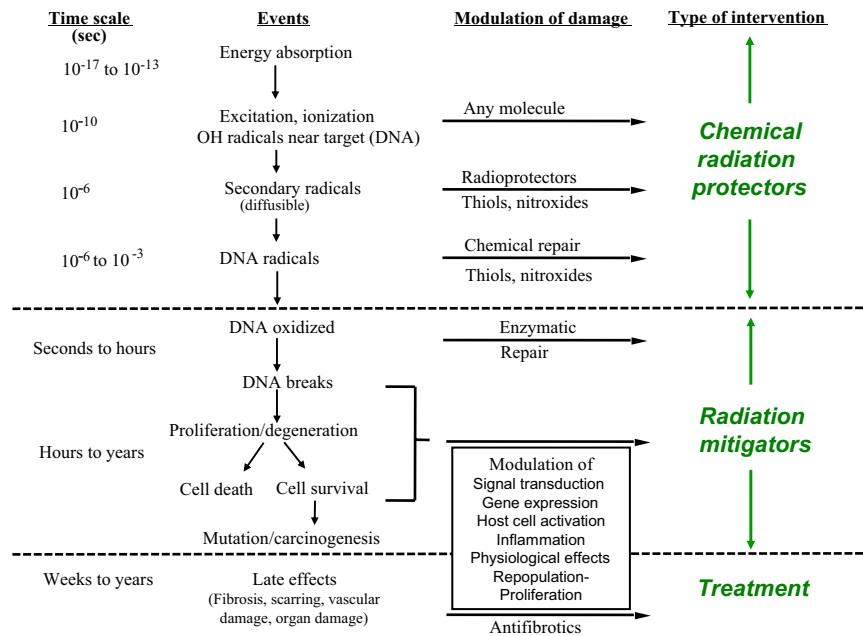


Figure 1. Sequence of events following radiation exposure. The chart is divided into three parts by dashed lines suggesting events and reactions that might be modified by radiation protectors (top), radiation mitigators, and treatment (bottom).

many free radical damage, the agent needs to be present at the time of radiation and in sufficient concentration to compete with radicals produced through radical-scavenging mechanisms. Many radical scavengers and antioxidants exist that can limit the oxidative stress induced by free radicals. Superoxide dismutase (SOD), catalase, glutathione peroxidase, and glutathione reductase are a few examples of naturally occurring antioxidants that defend against free radical-mediated damage, where the substrates are specific to each enzyme. General antioxidant defense is also provided by low molecular weight antioxidants, which are hydrogen atom-donating reducing agents such as ascorbic acid, tocopherols, polyphenols, and thiols such as glutathione. In this situation, the oxidants are neutralized by hydrogen atom donation, resulting in a less reactive or nonreactive product from the original oxidant and a radical product from the antioxidant, which no longer can exert detrimental effects.

Whereas radioprotectors need to have radical-scavenging properties and can also exert general antioxidant activity, all antioxidants cannot afford radioprotection [5]. This dichotomy may be a result of the relative reactivity of radiation-induced reactive species compared with those generated under conditions of general oxidative stress (i.e., H_2O_2 exposure). Scavenging hydroxyl radicals, such as those formed with radiation-induced damage, may be accomplished by almost any unsaturated organic molecule or molecule capable of H atom donation. Although hydroxyl radicals can be scavenged with equal efficiency by both ra-

dioprotectors and antioxidants, cellular and in vivo radioprotection is observed only with radioprotectors. This suggests that a secondary species is generated by hydroxyl radicals and is responsible for critical target (i.e., DNA) damage. This less reactive secondary species may not be scavenged by conventional antioxidants either because they do not accumulate in proximity to the secondary radical or they may not have kinetic reactivity to scavenge them effectively. Thus, thiols such as amifostine and the newly developed nitroxides have sufficient reactivity to efficiently scavenge secondary radicals. Conversely, well-known antioxidants such as vitamin C and vitamin E do not act as classic radioprotectors.

Amifostine: A Radioprotector in Use Clinically

Sulfhydryl compounds such as cysteine and cysteamine have long been known to act as radioprotectors via free radical scavenging and H atom donation [6, 7]. Since the initial description of sulfhydryl/thiol compounds as radioprotectors, more effective and less toxic agents have been described. Perhaps the best known agent in this class is amifostine. Other agents such as N-acetyl-L-cysteine and diethyldithiocarbamate have also been described as radioprotectors, although with lower efficacy at equitoxic doses in mice, compared with amifostine [8].

Amifostine is a phosphorothioate that is not taken into cells until it is dephosphorylated by alkaline phosphatase [9]. Once dephosphorylated, the agent freely diffuses into cells and can act as a free radical scavenger. Amifostine has

been shown to concentrate more rapidly in normal tissues than in tumor tissues in studies of tumor-bearing animals [10], which is thought to result from several factors, including the effects of tumor blood flow, the acidosis of tumors, and the lower expression of alkaline phosphatase. Additional potential mechanisms of protection have been described, including induction of hypoxia through increased oxygen use [11, 12] and condensation of DNA [13].

The radioprotective effects and selective concentration of amifostine into normal tissues led to extensive preclinical testing of the drug as a radioprotector and eventually to clinical testing. Amifostine has been evaluated as a radioprotector and chemoprotector in a large number of clinical trials, including a number of phase III trials. Randomized trials of amifostine as a radioprotector have been completed for patients with squamous cell carcinoma of the head and neck, non-small cell lung cancer, and pelvic malignancies. These trials have included the use of amifostine in the setting of radiation and chemoradiation, both in the definitive and adjuvant settings. In these studies, amifostine was evaluated as a mechanism to prevent or reduce acute and late xerostomia, mucositis, dysphagia, dermatitis, pneumonitis, proctitis, and cystitis. A number of series have also evaluated tumor control as an endpoint. Although many trials have been completed, many of the early reported series were small, used dosing schedules that varied markedly from one study to the next, and evaluated a heterogeneous group of patients.

More recently, amifostine was evaluated in additional randomized trials and was found to reduce toxicity when added to radiotherapy [14], leading the American Society of Clinical Oncology (ASCO) to state that amifostine may be considered for the prevention of xerostomia during fractionated radiotherapy [15, 16]. These guidelines state that current data do not support the use of amifostine in the setting of chemoradiotherapy, which has become standard therapy in a number of settings, especially in radiotherapy for advanced head and neck malignancies. Additionally, the 2008 ASCO guidelines state that the data are insufficient to recommend the use of amifostine to prevent mucositis associated with radiotherapy for head and neck malignancies or esophagitis associated with chemoradiotherapy for non-small cell lung cancer. Concerns about tumor protection and toxicity of the agent have led to controversy regarding the appropriate setting for its use [17].

Nitroxides: Promising Agents in Clinical Development

Amifostine is the only radioprotector currently in clinical use. A number of other compounds are in various stages along the pathway of clinical development as radiation protectors. Nitroxides are among the most promising agents for

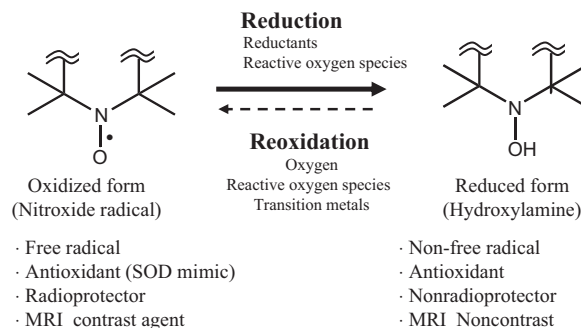


Figure 2. Schematic diagram of nitroxide radical conversion to hydroxylamine and chemical properties associated with both forms.

Abbreviations: MRI, magnetic resonance imaging; SOD, superoxide dismutase.

future use as radiation protectors. Although a number of these agents are useful in the laboratory as radiation protectors, not all have the requisite characteristics that allow them to be used clinically. We highlight the development of nitroxides as radioprotectors and describe the current status of their clinical development.

Our laboratory has shown that stable nitroxide free radicals and their one-electron reduction products, hydroxylamines, are recycling antioxidants that protect cells when exposed to oxidative stress, including superoxide and hydrogen peroxide (Fig. 2) [18]. Likewise, preclinical studies have shown that the oxidized form of a nitroxide is a radioprotector in both in vitro (cell survival) [19] and in vivo (lethal total body radiation) [20] models. Although the hydroxylamine exhibits antioxidant activity, it is incapable of protecting against radiation damage [19]. The lead compound from this class for radioprotection is tempol (4-hydroxy-2,2,6,6-tetramethylpiperidine-1-oxyl).

As with any radioprotector, there is concern that systemic administration might protect tumor as well as normal tissue. Therefore, initial preclinical studies focused on topical application of tempol with the anticipation that systemic levels of the drug would be low and hence not sufficient to protect tumor tissue. Preclinical studies in guinea pigs revealed that topical application was effective at preventing radiation-induced alopecia [21, 22]. A phase I clinical trial in patients receiving whole-brain radiotherapy suggested that tempol may be effective at preventing radiation-induced alopecia with only mild (grade I and II) toxicity [23]. Both the preclinical and clinical studies showed that systemic levels of tempol were negligible following topical application.

Subsequent preclinical studies determined if tempol was capable of radioprotection when administered systemically. Figure 3A shows that tempol protects against radiation-induced damage to salivary glands and does not alter

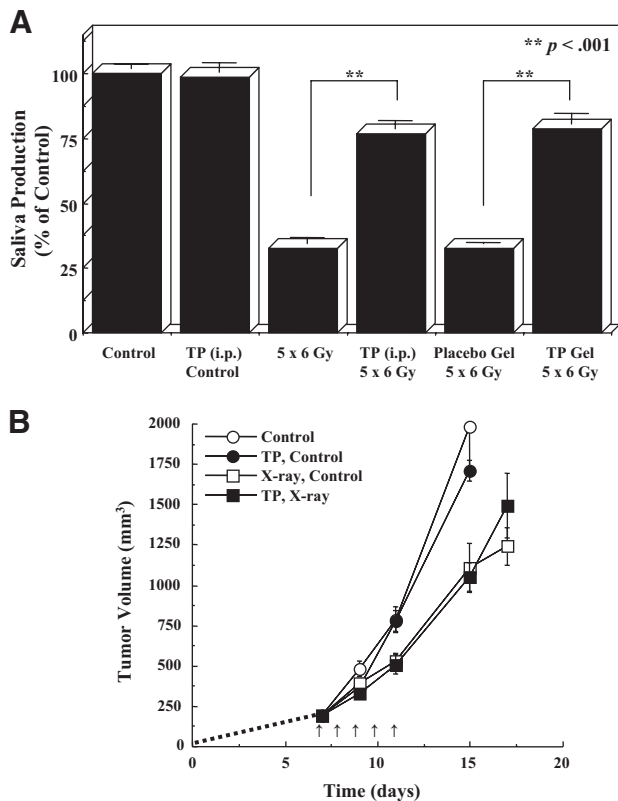


Figure 3. Mice were exposed to local fractionated radiation treatment to the salivary glands (A) or tumor-bearing leg (B) with and without systemic tempol (TP) administration (275 mg/kg given 10 minutes prior to each radiation fraction). Note that the same TP dose and timing were used in each study. (A): Saliva production 2 months postradiation for mice exposed to 5 daily fractions of 6 Gy. In addition to systemic delivery of TP, mice were also treated topically for 20 minutes prior to radiation with a TP gel formulation. (B): Radiation tumor (SCCVII) regrowth study for local fractionated radiation treatment with and without systemic TP administration. Tumors received 5 daily fractions (Monday to Friday) of 3 Gy.

Adapted from Cotrim AP, Hyodo F, Matsumoto K et al. Differential radiation protection of salivary glands versus tumor by Tempol with accompanying tissue assessment of Tempol by magnetic resonance imaging. *Clin Cancer Res* 2007;13:4928–4933, with permission.

tumor growth after irradiation (Fig. 3B), suggesting that delivery of the agent prior to irradiation would not alter tumor control [24]. A possible explanation for the apparent differential protection of normal as opposed to tumor tissue is shown in Figure 4. In the oxidized form, tempol is paramagnetic and provides T_1 contrast on magnetic resonance imaging (MRI) [25]. Because of this unique property, the active, radioprotective form of tempol can therefore be followed temporally using MRI. Tumors were grown on the neck region of a mouse that would allow a single MRI slice to include the tumor, salivary gland area, and normal leg

muscle (Fig. 4A, 4B). As shown in Figure 4C, tempol injection resulted in image enhancement, which decreased as a function of time after injection. The decrease in tempol MRI enhancement represents cellular reduction of tempol to the hydroxylamine tempol-H [26], which is nonradioprotective. By following the various regions of interest outlined in Figure 4B, a quantitative temporal assessment of tempol concentration in its radioprotective form in tissue can be determined as shown in Figure 4D. This plot shows that the rate of reduction of tempol is similar for normal muscle and salivary gland tissue; however, it is significantly faster in tumor tissue. Collectively, the data shown in Figures 3 and 4 are consistent with the hypothesis that differential radioprotection resides in a faster reduction to the nonradioprotective hydroxylamine in tumor than in normal tissue [24].

These preclinical findings provide feasibility to evaluate tempol as a radioprotector in clinical trials for cancer patients treated with radiation. Coupling MRI with such a trial would permit a novel dimension that could provide extremely important information with respect to the timing of tempol administration and radiation treatment. For example, before radiation treatment, a pilot tempol/MRI scan could be conducted to determine the tempol reduction rates in tumor and normal tissues encompassed in the proposed treatment field. Based on the reduction rates, the optimal timing of tempol administration with respect to radiation treatment to provide selective radioprotection to normal tissues could be determined. What is unique about this approach is that the therapeutic agent in this case (tempol) can be visualized by MRI. There are few therapeutic agents used in cancer management (excluding radiolabeled agents) that can be followed by non-invasive imaging. Before such an approach can be considered for clinical trials, more research is required to determine if tempol reduction rates in tissues change during fractionated radiation treatment. Lastly, monitoring the profiles of reduction/oxidation of nitroxide–hydroxylamine couples may actually serve as a viable approach to assess the global redox status in tissue using MRI and have potential applications in various disease states resulting from oxidative stress and inflammation.

Other Antioxidants

With the understanding that free radicals perpetuate a significant amount of the damage caused by ionizing radiation, multiple vitamin antioxidants have been tested as a method to reduce the toxicity of radiotherapy. Antioxidant compounds such as glutathione, lipoic acid, and the antioxidant vitamins A, C, and E have been evaluated in this context. A great deal of preclinical and clinical information has been

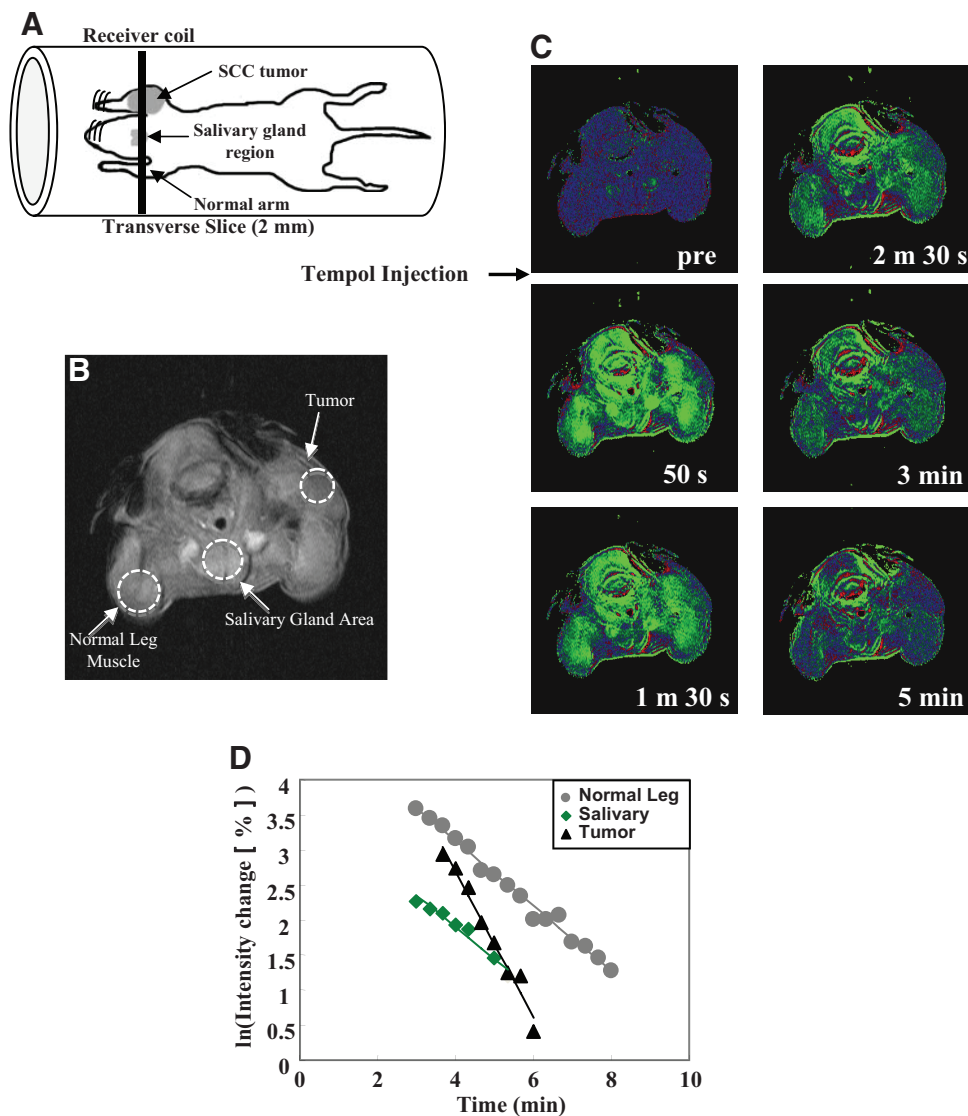


Figure 4. T₁-weighted MRI images using tempol. (A): Schematic diagram of the placement of a tumor-bearing mouse in a resonator and the MRI slice selected that includes normal muscle tissue, the salivary gland region, and the tumor in the contralateral leg. (B): T₂-weighted MR image of the selected slice before injecting tempol to ensure that the target tissues were in the field of view. (C): T₁-weighted images of the selected slice before injection of tempol and as a function of time after i.v. tempol injection. (D): Tempol reduction rates for the selected regions of interest shown in (B).

Abbreviations: MRI, magnetic resonance imaging; SCC, squamous cell carcinoma.

Adapted from Cotrim AP, Hyodo F, Matsumoto K et al. Differential radiation protection of salivary glands versus tumor by Tempol with accompanying tissue assessment of Tempol by magnetic resonance imaging. *Clin Cancer Res* 2007;13:4928–4933, with permission.

accumulated that describes the effects of combining radiotherapy with antioxidants. In general, the efficacy of these naturally occurring agents as radioprotectors is less than that for the synthetic agents previously described. It is important to briefly review the available literature on the radioprotective abilities of other available antioxidants and to understand the important implications of using these agents during the course of radiotherapy.

One of the major concerns with the use of supplemental nutritive antioxidants or other antioxidants during the

course of radiotherapy is the possibility of tumor protection through nonselective free radical scavenging. As described above for agents such as amifostine, selective uptake or activity in tumor tissue is essential to realize a gain in the therapeutic ratio.

A number of trials have been performed with antioxidants delivered during the course of radiotherapy, with the goal of reducing normal tissue toxicity, in many instances with promising results. For example, antioxidants have been delivered concurrently during the course of radiother-

apy to reduce xerostomia [27], mucositis [28, 29], pulmonary fibrosis [30], cystitis [31], and alopecia [32]. With this approach of delivering the antioxidants concurrently, tumor protection has been raised as a major concern [33].

Unfortunately, the use of antioxidant vitamins, such as alpha tocopherol and beta carotene, during the course of radiotherapy was associated with evidence of poorer tumor control in randomized trials [28, 34]. The lower toxicity associated with the use of these agents is appealing, but not at the cost of poorer tumor control. These findings reinforce the importance of preclinical testing of radioprotectors to verify a lack of tumor protection. Regardless of the extent of preclinical evidence supporting a lack of tumor protection, clinical trials testing new radioprotectors should carefully document tumor control. Topical application has been used to minimize the possibility of systemic absorption and interference with tumor response to radiation [32]; however, caution is advised because even topical applications for the prevention of mucositis in head and neck cancers have been associated with evidence of poorer tumor control [29].

When discussing antioxidants as radioprotectors, it is worth mentioning the use of SOD as a method to prevent radiotherapy-induced toxicity. Ionizing radiation results in the formation of superoxide radicals that are highly reactive and potentially damaging to cells. SOD is an enzyme that is naturally present in human cells. It catalyzes the conversion of superoxide to oxygen and hydrogen peroxide and functions as an antioxidant during normal conditions and after radiation.

Intracellular localization of SOD is critical to its effectiveness as an antioxidant; however, SOD is a large protein and does not freely enter into cells. To circumvent this limitation, much of the work evaluating SOD as a radioprotector has used gene therapy to increase the levels of SOD in tissues to be irradiated to prevent or decrease radiation-induced mucositis [35], esophagitis [36], pneumonitis [37–39], and fibrosis [40, 41] in animal models. Importantly, studies of tumor response after these delivery methods [39] and studies of tumor cell lines expressing various quantities of manganese SOD have been completed and suggest that this therapy does not decrease tumor response to radiation [42]. Additional work needs to be completed to determine if these findings can be successfully translated into clinical trials.

Nonantioxidant Radioprotectors

Efforts to reduce early or late toxicities of radiotherapy have led to the development of a number of agents that can be delivered before or at the time of radiotherapy to enhance the survival of critical cell compartments. These agents do

not fit the description of a chemical radioprotector but do prevent radiation-induced cell death and can thus be described as radioprotectors.

One example of such an agent is the hormone melatonin. Melatonin is thought to act as an antioxidant itself [43–45], but also acts to increase the expression of antioxidant enzymes such as SOD and glutathione peroxidase [46, 47]. Radioprotection with melatonin and melatonin analogs has been documented in a number of animal models [48–53]. Importantly, melatonin has also been shown to have direct antitumor effects [54] and has been described as a radiation sensitizer for tumors in animal models [55].

The use of melatonin as a radiation sensitizer for tumor cells and as a radioprotector for normal cells was tested clinically in a phase II Radiation Therapy Oncology Group trial [56]. In that study, patients were randomized to either morning or nighttime high-dose melatonin during radiotherapy. Melatonin was continued after radiotherapy until progression or until 6 months. Although melatonin delivered concurrently with radiation was well tolerated, there was no evidence that the treatment resulted in a longer survival time or better neurologic function than in historical controls.

Targeting signal transduction pathways has also been explored as a mechanism to protect organisms and tissues from ionizing radiation. One example of this approach is the use of the polypeptide CBLB502 [57], which binds to and activates Toll-like receptor 5 (TLR5), which is expressed in enterocytes [58] and intestinal endothelial cells [59]. Activation of TLR5 results in activation of nuclear factor κ B, which is thought to play an important role in the response of the intestine to irradiation [60]. Delivery of CBLB502 prior to and shortly after radiation protected mice and rhesus monkeys from lethal total body irradiation, with evidence of less damage to the intestine and bone marrow [57]. Importantly, no evidence of tumor protection from irradiation was evident. It is possible that this type of protector could be useful not only for accidental exposures but also for therapeutic radiation, when large areas of intestine or marrow could be considered dose limiting.

Radiation Mitigators

Radiation-induced late normal tissue toxicity is increasingly being appreciated as a phenomenon of ongoing changes in tissue after radiation but prior to the manifestation of toxicity. These events include ongoing mitotic cell death and perpetually active cytokine cascades that can lead to vascular damage, tissue hypoxia, and excessive extracellular matrix deposition [61]. Radiation mitigators aim to interrupt these cascades or intervene to prevent the perpet-

Table 1. Representative radiation mitigators

Class	Representative agent(s)	Proposed mechanism	Use	Clinical status [62]
Growth factor	Palifermin	Stimulates differentiation, proliferation, DNA repair, and detoxification of reactive oxygen species [78]	Mitigation of mucositis [16], mitigation of xerostomia [79]	In use to prevent or reduce mucositis after transplantation; ongoing testing with radiation, chemotherapy, or both to prevent mucositis
Protease inhibitor	Bowman-Birk proteinase inhibitor	DNA repair: increases fidelity of and stimulates DNA repair [63–65]	Chemopreventative [66], antifibrotic [67]	Ongoing testing as a chemopreventative
Dithiolthione	Oltipraz	Increased nonprotein sulfhydryl in cells: enhances radiation-inducible glutathione S-transferase and microsomal epoxide hydrolase expression	Chemopreventative, protector against total body exposures [68]	Ongoing testing as a chemopreventative
ACE inhibitor	Captopril, enalapril, ramipril	Inhibition of angiotensin II production, suppression of proliferation, inhibition of nitric oxide synthase, prevention of radiation-induced TGF- β , suppression of chronic oxidative stress, suppression of aldosterone [69]	Prevention of radiation-induced nephropathy [70], optic neuropathy [71], and pneumonitis [72]	Ongoing and recently completed clinical trials for prevention of radiation-induced nephropathy and pneumonitis; clinical data available suggesting stabilization of renal function in established radiation-induced nephropathy [69]
Isoflavone	Genistein	Hematopoietic stem cell quiescence [69], tyrosine kinase inhibitor [73], antioxidant	Chemopreventative, protector against total body exposures [74]	Ongoing testing as a chemopreventative, direct anticancer agent, radiation sensitizer in tumor tissue
Hmg-CoA reductase inhibitors (statins)	Simvastatin, pravastatin, lovastatin	Inhibition of the Rho/CCN2/extracellular matrix cascade [75]	Mitigation of radiation enteropathy [76], pulmonary fibrosis [77]	Ongoing trials testing as a mitigator of rectal injury with radiation, testing as a chemopreventative
COX2 inhibitors/NSAIDs	Celecoxib, aspirin, ibuprofen,	Inhibits COX2 activity in the setting of increased COX2 expression and prostaglandin synthesis after radiation [80]	Radiation sensitizer [81], chemopreventative [82], mitigator of mucositis	Ongoing trials as a chemopreventative, ongoing and completed trials as a radiation sensitizer for tumors, ongoing trials for prevention of mucositis
TGF- β signaling inhibitors	Halofuginone, ID11, SM16	Inhibits TGF- β signaling that results in activation of profibrotic pathways	Antifibrotic	Ongoing clinical trials in disease characterized by fibrosis

Abbreviations: ACE, angiotensin-converting enzyme; COX2, cyclooxygenase 2; Hmg-CoA, 3-hydroxy-3-methyl-glutaryl-coenzyme A; NSAIDs, nonsteroidal anti-inflammatory drugs; TGF, transforming growth factor.

uation of damage and thus reduce the expression of toxicity. Alternatively, radiation mitigators can be agents delivered during or shortly after exposure to repopulate a critical cell compartment such as the mucosa or bone marrow. In this instance, the mitigator is used to prevent acute toxicity. For radiologic terrorism and space research, much of the focus of mitigators has been in the field of developing chemopreventatives to reduce carcinogenesis of total body exposures. Table 1 summarizes several promising radiation mitigators [16, 62–82]. A few examples are discussed in detail below.

Many cytokines and growth factors are radiation mitigators when used near the time of radiation. These agents stimulate the differentiation of stem cells in bone marrow or the intestine, thus preventing bone marrow failure or gastrointestinal syndrome after total body exposure. A number of cytokines and growth factors have been explored as radioprotectors/mitigators. For example, G-CSF can effectively reduce the lethality of total body radiation exposure by assisting in marrow recovery [83, 84]. Recently, interest in keratinocyte growth factor

(KGF) as a possible mitigator has spurred both preclinical studies and clinical trials.

KGF is a growth factor that stimulates a number of cellular processes such as differentiation, proliferation, DNA repair, and detoxification of reactive oxygen species [78]. These properties make KGF an attractive method to stimulate the recovery of mucosa after ionizing radiation. Accordingly, delivery of KGF in animal models prevents radiation-induced xerostomia [79] and mucositis [85].

Palifermin is a recombinant human KGF that is approved for use in decreasing the incidence and duration of severe oral mucositis in patients with hematologic malignancies who receive high doses of chemotherapy and radiation therapy followed by stem cell rescue. The success of palifermin in patients with mucositis after cytotoxic therapy [86] led to attempts to evaluate its use in patients with head and neck cancers receiving chemoradiotherapy, in whom mucositis can be severe and prolonged.

Evaluation of palifermin in a phase II clinical trial as a method to reduce mucositis in patients receiving radi-

ation and chemotherapy for head and neck cancer did not find significantly less mucositis, dysphagia, or xerostomia with the agent than with placebo when evaluating the total study population; however, patients who received hyperfractionated radiotherapy had a lower incidence of mucositis and a shorter duration of mucositis [87]. A number of methodological problems led investigators to conclude that future studies should employ standardized tools for the assessment of mucositis, increase the duration of assessments to ensure that this period encompasses the resolution of mucositis in most patients, and use higher doses of palifermin. Because normal mucosa and squamous tumors may express the receptor for KGF, the investigators simultaneously reported the long-term follow-up of survival and progression-free survival outcomes in this study, showing that the delivery of palifermin did not influence disease control.

Mitigators of late radiation damage frequently target radiation fibrosis, a common and potentially debilitating complication of therapeutic radiotherapy. A variety of agents that protect against fibrosis have been evaluated as mitigators of radiation fibrosis. Transforming growth factor (TGF)- β plays a critical role in the development of radiation-induced fibrosis. It is therefore not surprising that many of the agents that have been used to prevent the development of radiation fibrosis directly or indirectly inhibit the TGF- β signaling pathway.

TGF- β receptor inhibition has shown the ability to prevent lung fibrosis after radiation exposure in animal models [88, 89]. An alternative mechanism is the use of halofuginone, a small molecule that inhibits TGF- β signaling, which has been shown in animal models to inhibit radiation-induced fibrosis [90]. Many of these agents are interesting for possible translation into the clinical setting; however, given the important role of TGF- β in tumor dormancy, progression, and metastasis [91], a thorough evaluation of tumor protection with these strategies is obviously important to ensure safe clinical translation.

CONCLUSIONS AND FUTURE DIRECTIONS

Radiation therapy is frequently used in the definitive management and palliative care of patients with cancer. Treatment of tumor tissue inevitably results in the irradiation of surrounding normal tissues. Acute and late radiation-induced normal tissue toxicity can have a significant impact on compliance with therapy and quality of life. Technological advances may result in lower normal tissue exposure, but it is expected that technologic approaches will not completely prevent toxicity in irradiated fields.

The use of compounds to protect normal tissues or minimize toxicity after damage has occurred may provide the

ability to reduce toxicity for patients treated with radiotherapy and may provide methods to treat individuals exposed to radiation through terrorism. Evidence of efficacy, lack of tumor protection, and acceptable toxicity are all important considerations for developing these agents. Amifostine remains the only agent currently in clinical use as a radioprotector. A number of other candidate compounds, such as tempol, will be tested in future years as a way to reduce radiation-induced normal tissue toxicity and complications.

Although prevention of radiation toxicity may provide the best opportunity to minimize impact on quality of life, few radioprotectors are in clinical use and the treatment of radiation injury remains an important mechanism to deal with radiation-induced toxicity. Antifibrotic treatments, such as pentoxifylline and vitamin E, have shown promise in clinical trials. Newer technologies, such as gene therapy, may offer the ability to reverse radiation-induced toxicities such as xerostomia [92]. Technologic improvements and the development of radioprotectors, mitigators, and treatments for toxicity are all important areas of research as methods for improving quality of life in patients who have received radiotherapy.

FINAL COMMENT

The distinguished clinician/scientist who is honored and recognized by this special issue, Dr. Eli Glatstein, is a true champion of translational research. In fact, he was practicing translational research long before the term became popular. While Eli never considered himself a bench scientist, he was constantly probing the radiation biology literature for novel approaches to treating cancer. His conviction to translate laboratory findings to clinical trials was influenced by two major figures in radiation oncology and biology, Henry Kaplan and Jack Fowler, under whom Eli trained. Eli often quoted Kaplan, "If you want to treat Hodgkin's disease you have to think like a Reed Sternberg cell," emphasizing the need and necessity of understanding the biology to effectively treat the cancer. From Jack Fowler, he acquired two fundamental traits, enthusiasm for research (and life) and the talent for diplomatically and effectively questioning established dogma. Eli does not shy away from addressing tough issues. In thought-provoking editorials over the years on a variety of topics, Eli has established himself as the conscience of the radiation oncology community.

Not only is Eli an accomplished experimental radiation oncologist, but he is also a well-rounded oncologist in general. Eli has participated in and contributed to countless rounds in surgery and medical oncology, positively influencing a younger generation of oncologists and crystallizing ideas for future translational studies involving multidisciplinary approaches to cancer treatment. Through-

out his career, Eli has consistently embraced the concept that an in-depth understanding of the biology of cancer is the correct path toward improving cancer treatment. His support and dedication to this concept is appreciated by all of those who have been fortunate to work with him.

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