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Treatment for Radiation-Induced Pulmonary Late Effects: Spoiled for Choice or Looking in the Wrong Direction?

Jacqueline P. Williams, Carl J. Johnston, and Jacob N. Finkelstein

Abstract

Due to the radiosensitivity of the lung, toxic endpoints, in the form of radiation pneumonitis and pulmonary fibrosis, are relatively frequent outcomes following radiation treatment of thoracic neoplasms. Because of the potential lethal nature of these normal tissue reactions, they not only lead to quality-of-life issues in survivors, but also are deemed dose-limiting and thereby compromise treatment. The mitigation and treatment of lung normal tissue late effects has therefore been the goal of many investigations; however, the complexity of both the organ itself and its response to injury has resulted in little success. Nonetheless, current technology allows us to propose likely targets that are either currently being researched or should be considered in future studies.

Keywords

Radiation; lung; pneumonitis; pulmonary fibrosis

General Background

Since the primary function of the lungs is to provide gaseous interchange, there is an innate requirement for the organ to be accessible to both the external and internal environments via inhalation and the circulation, respectively, leading to the evolutionary development of critical structural and physiological relationships between the air passages, respiratory parenchyma, and vascular system. However, an additional consequence is that the intimacy of these relationships together with the ease of access makes the lungs especially susceptible to a multitude of physical, chemical, and biological stressors that appear to be able to disrupt the delicate functional balance of this system with relative ease. Of course, through normal wound healing responses, such an injury may be completely resolved. However, depending on the source and severity, under many circumstances there is progression to a persistent, chronic pathology, which occurs through a complex cascade of processes, beginning with the acute injury and followed by an associated innate inflammatory response, culminating in abnormal remodeling and tissue repair [1].

With respect to this article, the pulmonary sequelae that are seen following *irradiation* are, indeed, varied and often long-lasting, and include edema, epithelial degeneration and subsequent regeneration, invasion of alveoli by the bronchial epithelium, endothelial sloughing, disruption of the microvasculature, and atelectasis [2,3]. Of clinical concern, the lung consequences, radiation pneumonitis and pulmonary fibrosis, that can develop in the normal tissue in the months to years after a standard course of radiotherapy obviously affect quality-of-life and may even be lethal in outcome, and are therefore recognized dose-limiting complications in the treatment of thoracic and related tumors [4-5], thereby, potentially, compromising cure. In addition, these deleterious downstream effects not only arise following localized high-dose pulmonary irradiation, but also are seen subsequent to the use of low-dose whole body irradiation used, for example, as part of preconditioning regimens for bone marrow transplantation [6,7]. As a result, the development of pharmaceutical agents that can protect

against, mitigate, or treat the development and expression of such morbid conditions has long been an aim for many investigators due to the potentially significant clinical benefit. Recently, this goal has gained further attention due to a perceived threat from nuclear or radiological terrorism, since accidental radiation exposures also have been shown to lead to the induction of these same lethal outcomes [8,9], indicating the need for a countermeasure against late effects in normal lung tissue for use following detonation of a dirty bomb or similar devices.

As demonstrated at both the bench and patient/victim level, the response to radiation injury in the lung is associated with a well-characterized progression: there is an apparent “delay” subsequent to the immediate injury (the so-called “latent period”), followed by an acute phase of alveolitis/pneumonitis, and a final late/chronic stage of pulmonary fibrosis [10,11]. A similar sequence of events is seen in the majority of mammals, including humans [12], which has allowed for the use of both small and large animal models in the study of dose-response relationships and the temporal development of the radiation-induced tissue injury. Such studies have spanned the majority of the last century and continue up until the present day [13-16] and yet, despite this breadth of data, the critical underlying mechanisms that lead to either radiation pneumonitis or pulmonary fibrosis remain elusive, thereby confounding the identification of agents that could effect a successful therapeutic strategy. This failure may be partly the result of the inherent limitations of extrapolating animal data to humans, but it also may reflect an incomplete comprehension of the available human data, which have suffered from a lack of standardized endpoints, limited follow-up, and patient and observer variability [14,17].

In this review, we will attempt to present an overview of our current understanding of the progression of events, both pathological and molecular, that are involved in the lungs’ response to radiation injury, identifying potential targets for intervention. This will include past and current areas of interest, although we hope to offer insight into some potential areas that may yet be explored.

Clinical Course of Radiation-Induced Lung Effects: Signs and Symptoms

Limiting the endpoints of interest to radiation-induced pneumonitis and pulmonary fibrosis, in the majority of patients who have received a fractionated high-dose course of external beam thoracic irradiation, an asymptomatic period that lasts about 1-3 months will occur following completion of therapy, after which time, radiation pneumonitis may begin to develop. The time to onset of the pneumonitis and its severity are interdependent on a number of treatment-related factors, such as the total dose delivered [18], the volume of lung irradiated [19,20], the fractionation schedule used [21,22], and the use of chemotherapy, particularly when administered concurrently [17,23,24]. Additionally, patient-related factors also affect outcome, such as pre-existing lung diseases [25], poor pulmonary function [25,26], and as yet unidentified genetic predispositions [27]. As a result of this broad array of contributing factors, the reported incidence figures for lung late effects vary significantly; for example, using the most sensitive techniques for detecting radiation pneumonitis, i.e. by radiologic means, incidence rates as high as 43% have been described [14], although many of these patients will be clinically asymptomatic. As a result, the incidence of symptomatic pneumonitis is considerably lower and is in the range of 5-15% of patients [14,15,28].

As suggested previously, this significant variability in reporting may result partially from the lack of a consistent clinical definition, since there is a broad spectrum of symptoms associated with pneumonitis that ranges from minimal and transient to fulminant. At the lower extreme, the chief symptom is a slight cough, although there may be a sensation of fullness in the chest; importantly, these signs may lead to a misdiagnosis or can be erroneously ascribed to other disorders, which may have led to an under-reporting of the true incidence rate [17]. At the opposite extreme, patients can present with severe respiratory insufficiency and cyanosis,

which may progress to acute cor pulmonale within days. Overall, within the clinical range, the principal symptoms of radiation pneumonitis, in order of prevalence, are dyspnea, a nonproductive cough, and a high, spiking fever that is, nonetheless, transient [11,14,29].

Although patients with acute pneumonitis may exhibit complete resolution of their signs and symptoms, unfortunately, the majority of them will go on to develop progressive pulmonary fibrosis; interestingly, this chronic condition also has been shown to occur in the absence of a preceding acute phase. In general, pulmonary fibrosis evolves between 6 to 24 months post-treatment, but then stabilizes after 2 years [29]. The condition can result in a chronic pulmonary insufficiency, although this will be dependent upon the volume of lung treated, since fibrosis, like radiation pneumonitis, is characteristically restricted in its appearance to within the portal field. Where a large volume has been irradiated, the chronic insufficiency may progress to chronic cor pulmonale from the resultant pulmonary hypertension and othorpnea, with associated cyanosis, hepatomegaly, or liver tenderness [14].

Pulmonary Response to Radiation Injury: Targets and Therapies

Although the weeks following irradiation are described clinically as the latent period, nonetheless, biologically, there is a cascade of events that begins immediately following injury and continues through to the full expression of the clinical syndromes. Indeed, within nanoseconds, radiation induces direct and/or indirect effects in the intra- and inter-cellular compartments of an organ or tissue through the induction of free radicals and the accompanying oxidative stress [30]. As a result, this event should and does resemble many other DNA-damaging injuries, so that the immediate pulmonary response to radiation injury is similar to that of many normal wound-healing responses. However, for reasons that are not yet clear, the lung injury induced specifically by radiation fails to fully repair and resolve (a result not specific to the lung, but also seen in many other normal tissues, as described elsewhere in this issue), so that the tissue enters a progressive and dysregulated process that culminates in the acute and/or late endpoints of pneumonitis and fibrosis. Classically, such radiation-induced injuries were deemed to be products of cell loss in either the parenchymal or vascular compartments; however, they are now seen as the result of a complex, orchestrated interaction between multiple cell types, initiated and perpetuated through inter- and intra-cell signaling [31,32].

In order to better understand the complex pulmonary response to injury and thus identify potential targets for intervention, the reader first must have a degree of comprehension of the normal lung physiology. Although there are approximately 40 different cell types in the lung [33], the majority of researchers in this area have focused on those cells that are intimately involved in the functional unit of the lung, i.e. the terminal bronchiole and the respiratory parenchyma that it serves: the respiratory bronchiole, the alveolar duct and sac [3,34]. At the cellular level, the alveolar ducts and alveoli are lined by a single layer of epithelial cells, the majority of which (>90%) are the squamous type I pneumocytes interspersed with the more cuboidal type II pneumocytes. In addition to being precursors to the type I pneumocytes, type II pneumocytes contain lamellar bodies that synthesize and secrete pulmonary surfactant; surfactant regulates the surface tension in the aqueous layer (alveolar hypophase) that lines the alveolar surface [35]. This epithelial cell layer is separated from the endothelium of the closely-associated capillaries by their respective basement membranes; in much of the alveolar wall, these membranes are fused, thus facilitating gas transfer. Even where the membranes are not fused, the stromal space is minimal, although it may still contain pericytes, fibroblasts (including myofibroblasts), smooth muscle, collagen, etc. [3]. In addition, macrophages may be present within the alveolar space; these are migratory cells from the bone marrow and provide a source to the lung of a considerable number of cytokines.

Other cells found in the lung that, to date, have received relatively little attention include mast cells and dendritic cells, although recent indications have suggested that these, indeed, may play important roles in radiation pathophysiology [36,37]. In general, however, it is the previously-mentioned cell types, specifically the inflammatory, fibroblastic, and epithelial cells, that appear to play the most critical roles in radiation-induced pulmonary pathogenesis [38], and it is therefore likely that it is these cells, their response to injury, and the participating signaling molecules that will provide us with the most promising targets for drug intervention.

Targeting Free Radical Production

As has been emphasized earlier, the progression to pulmonary late effects is temporally long and biologically complex, but nonetheless begins within nanoseconds of the injury with the induction of free radicals, the classically recognized mechanism for radiation's genotoxic effects. Efforts have long been made to differentially reduce the effect of such radicals in normal versus tumor tissue and, from decades of research, the only agent currently FDA-approved as a clinical radiation protector has emerged: the thiophosphate, amifostine (Ethyol®), whose active metabolite acts as a radical scavenger. Amifostine is approved for use as a cytoprotectant, relieving problems of dry mouth (xerostomia) in patients with head and neck cancer undergoing radiotherapy; however, it has proved less efficacious as a radioprotectant in other sites, with both positive [39,40] and negative [41] effectiveness demonstrated in the lung. This has been disappointing given the compelling basic research in rodent models that indicated that pretreatment with the drug should provide significant protection against late pulmonary toxicity [42,43]; the lack of efficacy may be a result of the drug's preferential uptake by alternate organs, such as the kidney and liver [44], as well as insufficient drug availability since toxicity limits both dose and scheduling.

Nonetheless, the role that free radicals play in radiation injury has meant that interest in the overall approach of anti-oxidant use has been maintained and has even increased in recent years because of the implications that, although reactive oxygen and nitrogen species play an effective role in innate immunity and cellular regulation, their release also may result in collateral damage in normal tissue [45]. Importantly, their chronic release has been proposed as a mechanism for a number of pulmonary distress syndromes, including those seen following irradiation [46,47]. Indeed, subsequent to the initial robust production of reactive oxygen species (ROS) [48], the chronic progression of events in radiation lung toxicity involves temporally cyclical, downstream patterns of inflammatory cell recruitment and cytokine generation [49,50], all of which can lead to the generation of further ROS. This link between oxidative stress and response has been hypothesized as being critical in both the development and progression of tissue deficits [51,52].

One approach to manipulating the chronic effect of ROS is to utilize the body's own natural enzyme defense system against oxidative damage, a system which includes the generation and activation of catalase, glutathione peroxidase and the superoxide dismutases (SOD) [53]. Preliminary work using glutathione has offered some encouraging results [51] and, indeed, investigators have shown an association between low glutathione peroxidase activities and susceptibility to pneumonitis [54], however, the majority of investigators have focused their attention on the SODs and their role in inflammation and related toxicities. For example, work from a University of Pittsburgh group has provided strong evidence that manganese superoxide dismutase (Mn-SOD, *SOD2*) expression is intimately involved in the generation of radiation resistance in a murine model for pulmonary sequelae [55] and that intratracheal introduction of the *SOD2* transgene can provide therapeutic, organ-specific radiation protection [56]. Although the enthusiasm for gene therapy per se has been diminished by recent events, nonetheless, the overall concept of targeting Mn-SOD is still being pursued by a number of investigators, using not only gene therapy, but also catalytic Mn porphyrin mimetics [57,58].

Other anti-oxidant approaches also have been investigated. For example, utilizing a SOD/catalase mimetic, Eukarion (EUK)-189, investigators have demonstrated protection in both intracellular and extracellular injury models [59,60], as well as in an *in vivo* rat lung model that indicated that continuous infusion of EUK-189 by Alzet pump reduced induction of micronuclei (marker of DNA damage), both immediately and, importantly, weeks following the initial injury [61]. An alternative approach, targeting RNS instead of ROS, has shown that both pre- and post-irradiation treatment with nitro-L-arginine methyl ester (L-NAME), a nitric oxide synthase (iNOS) inhibitor, provided protection against acute tissue damage in the lung, again measured by the number of micronuclei in fibroblasts [46].

Another interesting, although less specific, anti-oxidant approach that has provided some indications of success has been the use of genistein, a soy isoflavone that acts both as a non-specific protein kinase inhibitor and also as a ROS scavenger. Early studies in a lipopolysaccharide (LPS) lung injury model have shown that genistein lowers ROS levels, thereby reducing DNA damage [62]. More recent work has suggested that both single-dose pre-treatment and chronic post-radiation administration using genistein as part of a supplemented diet provides partial protection against the induction of radiation pneumonitis, as well as a reduction in the extent of fibrosis, although the mitigation was insufficient to prevent lethality in the high dose model [63,64]. Interestingly, since the treatment appears to reduce DNA damage (as assessed by micronuclei formation and 8-oxodeoxyguanosine measurements) [63,64], but has little effect on lipid (membrane) peroxidation, at least one of the groups has suggested that the failure to abrogate against lethality in this model may be due to differential targeting against intra- versus inter-cellular ROS production, i.e. administration of genistein is sufficient to reduce the relatively low levels of ROS induced by the intracellular damage, but is unable to counteract the chronic and accumulating levels due to the inflammatory response [64]. This suggests that dosing schedules may need to be tempered to the differential ROS induction, depending on the pathological point of progression. However, an alternative hypothesis, proposed by Cohen et al., is that such findings instead may cast doubt on the underlying premise that chronic oxidative stress plays any role in the development of late radiation-induced pneumonopathy [65].

Targeting Cell Death

The most fundamental effect of radiation on any tissue is to cause cell death; in general, this can occur within hours, through an apoptotic pathway, or following one or more cell divisions (mitotic catastrophe) and therefore may not be seen for weeks, dependent on the cell cycle time. Apoptosis has been identified experimentally in the lung within hours of injury [66]; although not definitively identified, the cell types most likely to be susceptible to this early death include the endothelial cells, the type II pneumocytes and, possibly, the alveolar macrophages. Although there have been few studies with respect to radiation, some research in other injury models have suggested that surfactant-associated protein (SP-A), a component of surfactant, protects against apoptosis [67]; interestingly, electron microscopy has indicated that the first pathophysiologic change can be seen in the type II pneumocytes as early as 1 hour post-irradiation in the form of depletion in the number of lamellar bodies [2], suggesting a protection mechanism. In addition, SP-A may be a regulatory factor of the burst of alveolar macrophages and neutrophils that are seen entering the lung following injury [68] and subsequent work by the same group has shown that tyrosine kinase and PI3K inhibitors block SP-A, suggesting that SPAR may be a receptor tyrosine kinase activated through the PI3K-PKB/Akt pathway [69]. Activation of this pathway, therefore, may help to explain the relative resistance of type II cells to injury and further promotion of their survival through this mechanism could assist in repopulation of the alveolar epithelium after irradiation.

Targeting Inflammatory Cell Recruitment

During the acute phase following radiation injury, inflammation is the predominant histological and physiologic feature, taking the form of macrophage infiltration into the air spaces and focal accumulations of mononuclear cells and mast cells [70,71]; these events can be accompanied by a decline in pulmonary function [72,73]. Interestingly, early studies in mouse lung models supported the assumption that chronic inflammation stimulates fibrosis development, and that this occurs as a result of the sequential release of cytokines and growth factors [49,50,74]. Such an assumption has supported the clinical approach to radiation pneumonitis of limiting the inflammatory reaction, particularly in light of the observed beneficial effects of glucocorticosteroids in many irradiated organs and tissues [75-77]. However, there is little evidence that such an intensive anti-inflammatory treatment, which mitigates the signs and symptoms of pneumonitis, has any effect on radiation fibrosis [78,79] and, of course, opens the patients up to the possibility of steroid-related adverse effects [80] as well as the late onset of pneumonitis during dose tapering [81].

Such limitations in steroid therapy have led some to investigate the use of non-steroidal anti-inflammatory drugs; for example, the frequent over-expression of cyclooxygenase-2 (COX-2) in lung cancers led many to assess the use of COX-2 inhibitors, such as Celecoxib, since there were indications that these induced both sensitization in tumors and protection against fibrosis in the normal tissue [82]. Unfortunately, to date, benefit from such approaches has been demonstrated in a few patients only [75,83] and even at the bench level, only limited success has been seen using anti-inflammatory agents such as indomethacin [84,85], so that groups such as our own have assessed the mitigating capacity of drugs with more pleiotropic attributes, such as the statins. Apart from their known HMG-CoA reductase inhibition activity, many of the effects of this family of drugs offer promise in the radiation mitigation field. For example, statins have been shown to improve endothelial cell function, a widely hypothesized target of radiation damage, possibly through the enhancement of eNOS transcripts [86]; many of the statins also have demonstrated antioxidant as well as immunomodulatory activity [87,88]. However, statins also have demonstrated a significant anti-inflammatory role in a number of diseases, for example in atherosclerosis, where they have been shown to decrease the risk of coronary heart disease by lowering levels of C-reactive protein [89]. Our group therefore examined the efficacy of one of the leading statins, lovastatin, as a mitigator against radiation lung toxicity since it has been shown to be a non-specific inhibitor of monocyte chemoattractant protein-1 expression, a chemokine that plays a critical role in macrophage migration and that we have shown is upregulated in the lung at 8-weeks post-radiation in a mouse model [90]. We subsequently demonstrated that, whether treatment was administered continuously following irradiation or delayed until 8 weeks, lovastatin was effective at both improving survival in a high-dose lung mouse model and was associated with a reduction in the appearance of pneumonitis and fewer infiltrating macrophages [91]. Since other statins also have demonstrated effectiveness in reducing pulmonary fibrosis in a number of different models [92-94], this is a class of agent that may be worth investigating further.

In addition to macrophage infiltration, neutrophil trafficking into the lung is also seen following many stimuli, including radiation, and we have indeed demonstrated that neutrophils are present in the lung within hours of irradiation [66]. Interestingly, other groups have shown that there is a differential neutrophilic and mast cell activation as a result of radiation injury that varies between mouse strains, suggesting that genetic predisposition may be a component of this response [71,95-97]. Currently, however, the role that neutrophils may play in the initiation and progression of pulmonary events is far from clear, although there is indirect evidence that they may serve as a useful target since treatment with pentoxifylline and dexamethasone, which induces a significant alteration in neutrophil numbers, has been shown to ameliorate

histological changes in irradiated lungs [98]. Nonetheless, it appears too early to suggest that agents targeted against neutrophils may be of use in radiation pneumonopathy.

Targeting Cytokine and Growth Factor Expression

In conjunction with, and often related to, the alterations in physiology, irradiation injury induces changes in the expression levels of cytokines [49,99,100], chemokines [90,101], prostaglandins [102,103] and adhesion molecules [104,105], and the roles that these various families of molecules may play in late effect progression are currently being heavily investigated. For example, the early response cytokines, tumor necrosis factor (TNF)- α and the interleukins (IL)-1 α , IL-1 β and IL-6, are all potent proinflammatory cytokines that have been shown to trigger inflammatory cell recruitment following pulmonary irradiation, infection, and inhaled toxicants [52,106-109]; these and other cytokines and growth factors, such as transforming growth factor- β (TGF β) and the fibroblast growth factors, that appear to play critical roles in radiation-induced late effect progression may provide us with significant pharmacological targets; these will be discussed only briefly here.

Tumor necrosis factor (TNF)- α , one of the early response cytokines, has been shown by many investigators to be upregulated at critical times, both early and late, during the lung's response to radiation [52,99,110]; in murine models, this response varies temporally and quantitatively between different strains, with one group suggesting that its expression may be critical in radiation-induced pneumonitis [31]. However, few attempts appear to have been made to date to target TNF- α as part of a mitigating strategy, although two studies have suggested that some recently-described radioprotective agents may be functioning through a TNF-related pathway [111,112].

Many investigators have suggested an important role for members of the *IL1* family in the development of both acute and chronic inflammation as part of a broad spectrum of conditions and diseases [113-115]. For example, it has been shown that localized expression of these proinflammatory cytokines leads to an influx of leukocytes and neutrophils at a site of injury or infection [116,117] and in addition, there have been recent demonstrations of IL1's ability to elicit strong profibrotic responses. For example, IL-1 β has been shown to be present in tissues that demonstrate myofibroblast accumulation and matrix deposition [118,119]. Specifically in the lung, animal radiation models have shown that IL-1 is elevated in the bronchoalveolar lavage (BAL) [120], supporting data from clinical studies that have demonstrated elevated IL-1 in the BAL and alveolar macrophages of patients with respiratory diseases [121-123]. Significantly, studies in animal models of pulmonary fibrosis have demonstrated that administration of an IL-1 receptor antagonist at the time of insult attenuates the fibrotic response [124].

Although these data suggest a critical role for *IL1* in the progression of fibrogenic disease, a role for *IL1* in the specific development of radiation-induced injury is yet to be firmly established. Earlier work from our group demonstrated significant increases in IL-1 β expression immediately following radiation injury [99] as well as persistence in IL1 expression through the development of late fibrosis [49,99]. Importantly, we also have presented clinical evidence indicating a predictive role for IL-1 α in patients developing pneumonitis subsequent to lung irradiation [125,126]. These findings suggest that it is not unreasonable to hypothesize that the use of agents such as anakinra (Kineret®), a commercially available IL-1 receptor antagonist, currently indicated in the treatment of a variety of inflammatory diseases [127-129], may mitigate both the early and late pulmonary responses and, indeed, bench-level studies in this area are currently ongoing.

Another of the early response genes that is seen both acutely and late following radiation injury is transforming growth factor (TGF)- β . Like the interleukins, this is a highly pleiotropic

molecule, although its functions can be broadly categorized into 3 main activities: TGF- β is a key cytokine in the regulation and inhibition of cell growth; it exerts immunosuppressive influences; it regulates the deposition of extracellular matrix components [130]. Due to the first two of these functions, the early expression of TGF- β that is seen during the immediate period post-radiation may be part of a homeostatic mechanism as the tissue attempts to self-regulate the wound-healing response. Interestingly, the development and persistence of high levels of plasma TGF- β during the course of pulmonary radiation therapy have been suggested by some to be predictive of late effects [20,131] and recent work has suggested an association between TGF- β single nucleotide polymorphisms and the risk of pneumonitis [132], although the correlation between TGF- β and susceptibility to lung late effects has not been seen by all groups [125,133]. In contrast, TGF- β likely plays a critical role in the final fibrosis phase, which is the culmination of progressive deposition of extracellular matrix material resulting in the loss of alveolar architecture [130]. This has led to a number of studies looking at the efficacy of inhibitors of TGF- β , and recent preliminary work using a small molecule inhibitor of TGF- β RI, SM16, has been shown to reduce the extent of radiation-induced lung injury in a rat model [134].

Miscellaneous Targets

Given the drumbeat of described complexity in this system, it is not surprising that, serendipitously, agents may have been found for which there appear no clear explanation for their efficacy in reducing lung late effects. A primary example of this is the observation of a mitigating effect in the lung by agents that inhibit activity in the renin-angiotensin system (RAS) [135-137]. RAS is a complex, blood-borne hormonal system in which the substrate (angiotensin) and enzyme (renin) are released into the circulation by the liver and kidney, respectively, although it should be noted that the most active peptide product in this system, angiotensin II, can be generated both within and independently of RAS [32]. Although RAS plays a major role in the short- and long-term regulation of arterial blood pressure and hence RAS inhibitors are used as anti-hypertensives, RAS has also been shown to stimulate vascular smooth muscle cell migration, proliferation and extracellular matrix deposition [138], enhance the expression of such proteins as MCP-1, intracellular adhesion molecule (ICAM)-1 [139], integrins and osteopontin [140], as well as enhancing the migration of inflammatory cells through the upregulation of inflammatory cytokines and chemokines [141,142]. As a consequence of this broad spectrum of activity, the successful administration of angiotensin converting enzyme (ACE) inhibitors in lung radiation models [136,137] may be a result of the targeting of the oxidant, inflammatory and/or fibrogenic pathways leading to radiation pneumonopathy. Interestingly, ACE inhibitors have also been identified as providing considerable mitigation against radiation-induced nephropathy [143,144], although the mechanism in this tissue may be different since, unlike in radiation nephropathy, cessation of ACE inhibitor treatment is followed by a rapid deterioration in lung function [32].

One last target that may need further investigation is that of the immune system, more specifically the alteration in immune responses that are seen in the lung following injury that may play a critical role in late effect progression. Indications of an immune role were suggested by early work in murine lung radiation models that showed that different strains exhibited different endpoints and progression [70,145]; differential responses were particularly seen with respect to Th1- versus Th2-type responses [146]. Recent work has indicated that the lung's response to bacterial challenge (endotoxin or flagellin) is through toll-like receptors (TLRs), TLR-5 and possibly TLR-4, and that these are differentially expressed in different mouse strains (and across the human population) and are associated with radiation resistance [147, 148]. It has therefore been suggested by some that activation of the innate immune system may provide radiation protection [149]; this approach is being actively pursued with respect to radiation countermeasures [150] and may have considerable therapeutic implications.

Concluding Remarks

The sequential pathologic events that occur during the development of pulmonary late effects appear to be well characterized and there is a broad spectrum of cells and signals that offer themselves as potential targets for mitigating late pulmonary injury, with the prospect of anti-oxidants, anti-inflammatories, anti-fibrotics, or specific anti-cytokines providing some degree of benefit. Unfortunately, the inherent complexity of the progression of events suggests that neither (i) a simple single target/agent nor (ii) an acute dosing schedule are likely to prove successful as mitigating strategies. By gaining a deeper understanding of the pathophysiology of pulmonary late effect progression, together with the temporal expression of the panoply of signals that are inherent to the process, it is hoped that we will eventually be able to develop a series of strategies that will intelligently target critical events and processes and thereby reduce or even abrogate these lethal diseases. However, an important consideration in all of this work is that the use of any treatment in the context of cancer radiation therapy (versus agents used for terrorism countermeasures) will need to be carefully assessed with respect to its effects on tumors, since there is the requirement for a differential to exist between normal versus tumor tissue in order to result in an improvement in the therapeutic ratio.

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