Significant variability exists in normal tissue reactions in patients with cancer receiving radiotherapy, with a subpopulation exhibiting increased toxicity to ionizing radiation. Genomic studies have proposed that single nucleotide polymorphisms in DNA repair genes, cytokines, and reactive oxygen species may play a role in clinical radiosensitivity. Additional research examining the association between genetic variants and radiation-induced inflammation and fibrosis may spur the development of targeted therapy in radiation oncology, which could increase cure rates and limit toxicity. As more people become long-term cancer survivors, oncology nurses must aggressively assess and manage late treatment side effects to optimize patient functioning and quality of life. The purpose of the current article is to describe the effect of ionizing radiation on normal and irradiated tissue, discuss genetic mutations that are proposed to influence radiosensitivity, and identify future areas of research on the association between genetics and radiation toxicity.

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The field of radiation oncology has undergone many advances. New technology has spurred the development of intensity-modulated and image-guided radiation therapy, proton therapy, high-dose rate brachytherapy, and stereotactic radiosurgery techniques (e.g., gamma knife, CyberKnife®). About 60% of all patients with cancer receive radiation therapy for curative intent, tumor control, or palliation of symptoms (Halperin, Wazer, Perez, & Brady, 2013). The goal of treatment delivery is to provide a precise dose of radiation to the tumor and limit damage to surrounding normal tissue. Although most patients tolerate treatment with minimal side effects, a subset of individuals develop severe toxicities as sequelae of radiation therapy. Molecular profiling of tumors has begun to revolutionize the systemic treatment of cancer, but radiation oncology lags in identifying genetic factors that may confer individual susceptibility to radiation injury and toxicity.

Radiosensitivity is influenced by the effects of ionizing radiation on intracellular DNA, leading to cellular damage or death via double-strand breaks. Radiation also triggers the release of multiple cytokines, which are regulatory proteins that exert their intracellular effects via receptors on immunomodulatory cells (Martin, Lefaix, & Delanian, 2000). About 5%–10% of patients who receive radiation therapy exhibit a heightened sensitivity to conventional radiation doses (Gatti, 2001; Oezsahin et al., 2005; Popanda, Marquardt, Chang-Claude, & Schmezer, 2009). To limit toxicity, standardized dosing regimens have been developed and extensively researched for safety and efficacy. Advances in genetic research would enable radiation oncologists to design personalized therapy and optimize treatment plans for each patient, which would increase efficacy and minimize acute and late side effects (Ghazali, Shaw, Rogers, & Risk, 2012; Henríquez-Hernández et al., 2012). The current article describes the effect of ionizing radiation on normal and irradiated tissue, discusses genetic mutations that are proposed to influence radiosensitivity, and identifies future areas of research on the association between genetics and radiation toxicity.
Pathophysiology of Radiation-Induced Tissue Damage

Radiation-induced inflammation and subsequent fibrosis are initiated by mediators such as interleukin-1, interleukin-6, and tumor necrosis factor alpha (TNF-α), which are produced from activated monocytes, macrophages, endothelial cells, and fibroblasts. Profibrotic proteins, such as transforming growth factor beta (TGF-β) and connective tissue growth factor (CTGF), control fibroblast production, collagen growth, and deposition of extracellular matrix. Their actions are antagonized by TNF-α and interferon gamma (Leask & Abraham, 2004; Yarnold & Brotons, 2010) (see Table 1). Interferon gamma, macrophages, and TNF-α are initiated by mediators such as interleukin-1, interleukin-6, and tumor necrosis factor-alpha (TNF-α), which are produced from activated monocytes, macrophages, endothelial cells, and fibroblasts. Profibrotic proteins, such as transforming growth factor beta (TGF-β) and connective tissue growth factor (CTGF), control fibroblast production, collagen growth, and deposition of extracellular matrix. Their actions are antagonized by TNF-α and interferon gamma (Leask & Abraham, 2004; Yarnold & Brotons, 2010) (see Table 1). In irradiated tissue, these proliferative and remodeling processes are dynamic and promote the development of a chronic fibrotic state in skin, soft tissue, blood vessels, muscles, and organs in the targeted region.

Radiation-induced fibrosis can lead to clinical toxicities that include pain, altered cosmesis, limited range of motion, decreased functional capacity of solid organs and structures, fistulas, and obstructions of hollow organs (see Figure 1). To limit morbidity, radiation oncologists have sought to identify radiosensitive patients prior to treatment. Researchers have proposed that genetic factors confer an increased risk of developing treatment-related toxicities, and the completion of the Human Genome Project in 2003 has accelerated the understanding of the association between genetics and human radiosensitivity (Barnett et al., 2009). The most common mutation identified in candidate gene and genome-wide association studies is single nucleotide polymorphisms (SNPs), a DNA sequence variation in which one of the nucleotides (i.e., adenine, cytosine, guanine, or thymine) may be ataxia-telangiectasia carriers and at risk of morbidity or mortality from radiation exposure (Swift, 1994).

Prior to the initiation of the Human Genome Project in 1990, researchers used the candidate gene approach to investigate genetic variants in families who appeared to have an inherited pattern of disease (Barnett et al., 2009). Candidate gene studies are based on existing information about a gene and its potential biologic or functional impact on a disease; the researcher also must have knowledge of the pathophysiology of the disorder being studied (Rosenstein, 2011). This research led to the discovery of the BRCA1 and BRCA2 genes, as well as the mutation that causes Lynch syndrome (hereditary nonpolyposis colorectal cancer). However, limitations exist in the candidate gene approach. The approach assumes that the researchers' hypothesis is correct about the association between a selected gene and its function, and only a single gene is examined. However, most disease states are proposed to be influenced in a polygenic fashion (Andreassen, Alsner, & Overgaard, 2002; Popanda et al., 2009).

Genome-wide association studies use microarray technology to scan thousands of gene sequences simultaneously without prior knowledge of a gene's position or function. Samples then are analyzed to identify markers that are commonly found in affected individuals, which enables researchers to examine the influence of multiple genetic variants on common and complex disorders (Parliament & Murray, 2010). Genome-wide association studies are the cornerstone of research to eliminate false-positive findings, determine causative agents, and design clinically relevant translational research projects (Masny, Jenkins, & Calzone, 2010). The first genome-wide association studies in oncology were performed on prostate, breast, and colorectal cancer populations in 2007.

The most common mutation identified in candidate gene and genome-wide association studies is single nucleotide polymorphisms (SNPs), a DNA sequence variation in which one of the nucleotides (i.e., adenine, cytosine, guanine, or thymine) is altered.

<table>
<thead>
<tr>
<th>Cytokine</th>
<th>Cellular Origin</th>
<th>Mechanism of Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>IL-1</td>
<td>Macrophages, monocytes, B cells, other cells</td>
<td>Induces interferon-γ production and mobilization of phagocytes, lymphocytes, and other immunomodulatory cells</td>
</tr>
<tr>
<td>IL-6</td>
<td>T cells, macrophages</td>
<td>Promotes inflammation by mobilization of lymphocytes and monocytes; mediates differentiation of B and T cells; inhibits inflammation by down regulation of TNF-α and IL-1</td>
</tr>
<tr>
<td>Interferon-γ</td>
<td>Natural killer cells</td>
<td>Activation of macrophages and other phagocytic cells; suppresses collagen formation</td>
</tr>
<tr>
<td>TGF-β1</td>
<td>T cells</td>
<td>Inhibits interleukin, interferon-γ, and TNF-α production; promotes anti-inflammatory effects</td>
</tr>
<tr>
<td>TNF-α</td>
<td>Macrophages, natural killer cells, endothelial cells</td>
<td>Mediates apoptosis, coagulation, increased vascular permeability, and prostaglandin production; promotes extracellular matrix degradation</td>
</tr>
</tbody>
</table>

IL-1—interleukin-1; IL-6—interleukin-6; TGF-β1—transforming growth factor beta 1; TNF-α—tumor necrosis factor alpha

Note. Based on information from Guerra et al., 2012; Leask & Abraham, 2004; Martin et al., 2000; Yarnold & Brotons, 2010.
is altered. The human genome is virtually identical among individuals, with about 0.1% of genetic differences from SNPs (Parliament & Murray, 2010). Most SNPs are low-penetrance, harmless changes in DNA composition that make each individual unique, as compared to highly penetrant variants that are rare but cause clinical symptoms of disease.

In candidate gene and genome-wide association studies, researchers have identified several SNPs that are proposed to play a role in radiation sensitivity by damaging DNA recognition or repair, promoting radiation-induced inflammation and fibrosis, or affecting scavenging of reactive oxygen species (Fernet & Hall, 2008; West, Dunning, & Rosenstein, 2012).

**Genetic Mutations and Double-Strand DNA Repair**

DNA can be damaged by multiple agents, including ionizing radiation affecting one or both helical strands. When damaged, DNA can be repaired, enter a state of dormancy (senescence), continue to divide without regulation, or undergo programmed cell death (apoptosis). The ATM gene is responsible for generating proteins to repair DNA and regulating replication until corrected (Andreassen et al., 2002). SNPs of this gene are responsible for the generalized radiosensitivity of those affected with ataxia-telangiectasia, but ATM mutations also have been associated with toxicity in patients with breast, prostate, and lung cancers who have been treated with radiation therapy (Gatti, 2001; Rosenstein, 2011).

In patients with prostate cancer treated with high-dose external beam radiation therapy, a higher incidence of proctitis and cystitis were associated with ATM mutations (Hall et al., 1998). In Cesaretti et al.’s (2005) study, ATM mutations were associated with an increased rate of rectal bleeding and erectile dysfunction in patients undergoing brachytherapy. However, two subsequent studies failed to find a link between ATM SNPs and radiotoxicity in patients with prostate cancer (Damaraju et al., 2006; Meyer et al., 2007). Conflicting results also were noted in patients with breast cancer treated with radiation therapy, with studies associating variant ATM with the development of fibrosis and other subcutaneous tissue effects (Ho et al., 2007; Ianuzzi, Atencio, Green, Stock, & Rosenstein, 2002). However, other studies reported no increased risk of early or late skin toxicities (Andreassen, Alsner, Overgaard, Sorensen, & Overgaard, 2006; Bremer et al., 2003). All of those studies were performed using candidate gene SNPs and were not replicated in subsequent research on large patient cohorts (Parliament & Murray, 2010; Rosenstein, 2011).

**XRCC genes** also are proposed to function in single- and double-strand DNA damage caused by radiotherapy. SNPs of XRCC genes have been investigated in candidate gene studies for adverse effects, with XRCCI implicated for late fibrosis and telangiectasia in patients with breast cancer treated in radiation therapy (Andreassen, Alsner, Overgaard, & Overgaard, 2003), but a large replication study noted no correlation (Andreassen et al., 2006). In patients with head-and-neck cancer treated with radiation therapy, XRCCI mutations have been associated with acute mucositis and dermatitis (Pratesi et al., 2011) and XRCC3 mutations with the development of severe dysphagia after intensity-modulated radiation therapy (Werbrouck et al., 2009).

**Single Nucleotide Polymorphisms of Pro-Inflammatory and Profibrotic Agents**

Several cytokines and growth factors are proposed to cause molecular changes in target tissues. TGF-β is a multifunctional cytokine that plays a vital role in radiation-induced inflammation and fibrosis. Ionizing radiation stimulates production of the polypeptide TGF-β1 from various epithelial and endothelial cells, with activated TGF-β1 increasing synthesis of extracellular matrix components while inhibiting proteolytic activity (Leask & Abraham, 2004; Yarnold & Brotons, 2010).

SNPs of TGF-β1 have been implicated in the development of radiation-induced fibrosis in several cancers, including head and neck, endometrial, cervical, and lung. In patients with head-and-neck cancer, polymorphisms of TGF-β1 were associated with a lower grade of skin fibrosis (Alsbeih, Al-Harbi, Al-Hadyan, El-Sebaie, & Al-Rajhi, 2010) but with a significantly greater risk of developing osteoradionecrosis in another study (Lyons et al., 2012). Variants of the TGF-β1 gene also have been correlated with late side effects in patients with endometrial and cervical cancers (De Ruyck et al., 2006). In patients with lung cancer treated with thoracic radiation alone or with concurrent chemoradiation, a lower risk of radiation pneumonitis was found in those with a TGF-β1 mutation (Yuan et al., 2009) and with an increased incidence of radiation esophagitis in another study (Guerra et al., 2012). A meta-analysis of 2,782 patients with breast cancer treated with radiation therapy reported no association between TGF-β1 SNPs and late radiation fibrosis (Barnett et al., 2012).
Single Nucleotide Polymorphisms of Reactive Oxygen Species

Reactive oxygen species are oxygen-containing molecules and are essential in mediating apoptosis, cellular homeostasis, and signaling, and they are a byproduct of mitochondrial transport during cellular respiration. Ionizing radiation increases intracellular reactive oxygen species production and causes damage to lipids, protein, and DNA in a process known as oxidative stress (Ghazali et al., 2012; Seong et al., 2010). Cells contain multiple substances (e.g., uric acid, vitamins C and E) and enzymes (e.g., catalase, superoxide dismutase) to prevent oxidative stress. Superoxide dismutase exerts substantial anti-inflammatory and antioxidant activity and may reduce fibrosis by preventing the conversion of fibroblasts to myofibroblasts (Yarnold & Brotons, 2010). Superoxide dismutase has been studied for its radioprotective effects, with animal models demonstrating decreased damage to lung and parotid tissue (Andreassen, 2005).

SNPs of superoxide dismutase have been examined in candidate gene studies for an association with radiation sensitivity and, like previous studies of other SNPs, have yielded conflicting results. Research on superoxide dismutase mutations have been performed primarily in patients with breast cancer who have been treated with radiation therapy, with most studies showing no association between superoxide dismutase variants and an increased risk of fibrosis, acute skin toxicity, or telangiectasia (Ahn et al., 2006; Andreassen et al., 2005, 2006).

Implications for Practice

- Educate patients on the increasing role of genomics in treatment decision making.
- Possess current knowledge of genomic research to be aware of conflicting data on the influence of genetic variants on radiosensitivity.
- Provide thorough assessment and intervention of patients with radiation-induced toxicities to improve outcomes and functional status.

Conclusions

Although radiogenomics is in its infancy, candidate gene and genome-wide association studies have identified several SNPs that are hypothesized to confer a higher risk of radiation toxicity. However, much of the research has yielded conflicting results or has been underpowered and not replicated in larger independent studies. Radiation oncologists can employ differing fractionation schedules and dose-volume variations to influence the development, timing, and severity of radiation therapy-mediated toxicity (Ghazali et al., 2012). In the past, a lack of consensus occurred on the methodology used to collect toxicity data and which grading scales should be selected (Alsner et al., 2008; West et al., 2012). Researchers also must consider the development of side effects that occur several years after completing radiation treatment to determine frequency and duration of data collection (Popanda et al., 2009). To address these concerns, the Radiogenomics Consortium was established in 2009 to perform additional research by collecting tissue samples, treatment plans, standardized toxicity scores, and patient outcome data from thousands of individuals treated with radiation therapy to create a database for future research (Barnett et al., 2012).

Patient variables that may influence individual response to radiation therapy include age, performance status, comorbidities, environmental exposures, and concurrent administration of chemotherapy (Andreassen et al., 2005). Genomics has the potential to shape personalized treatment in patients with cancer. Predictive assays would create a genetic risk profile to guide decision making by patients and healthcare providers. Proposed biomarkers of radiation sensitivity have focused on the role of T cells in repairing radiation-induced double-strand breaks. Studies using peripheral blood lymphocytes have determined that radiation toxicity is associated with decreased apoptosis in these cells, leading to delayed repair in irradiated normal tissue (Ozsahin et al., 2005; Schnarr, Boreham, Sathya, Julian, & Dayes, 2009). Additional research is needed to develop assays that are inexpensive, have high predictive value, and are generalizable to multiple cancer populations.

Various radioprotective agents have been used in clinical practice (e.g., amifostine, pentoxifyline in combination with vitamin E), but results have been mixed with reduction of fibrosis noted in some studies and refuted in others (Delanian, Porcher, Rudant, & Lefaix, 2005; Hensley et al., 2009). Researchers should work to develop pharmacologic and nonpharmacologic strategies to treat radiation-induced inflammation and fibrosis.

References


