REPORT

RADIATION-ENHANCED IMMUNE RESPONSE TO CANCER: WORKSHOP, ANAHEIM, CA, APRIL 17, 2005

ROBERT M. SUTHERLAND, PH.D.
Varian Biosynergy, Varian Medical Systems, Mountain View, CA

INTRODUCTION

There is rapidly growing interest in the potential for synergistic, clinically relevant therapeutic responses by combining radiation therapy with immune enhancement. Because of this interest, a small group of active researchers in this field convened in a workshop to summarize recent preclinical research on combining radiation therapy with immunotherapy and to assess implications for translational research in humans. This first meeting developed some concepts and directions that might help to guide the development of radiation-enhanced immune responses in the clinic. The focus of the workshop was primarily on enhancement of active T cell–mediated antitumor immunity through vaccination approaches, although we acknowledge that there are other mechanisms by which the immune system can effectively react to tumors, under specific circumstances, and that radiolabeled monoclonal antibodies and adoptive transfer of immune cells are additional promising therapeutic strategies.

Participants presented recent research data. Plans for future directions were also discussed. Among the results presented were data showing that stimulation of in vivo proliferation and differentiation of dendritic cells with cytokines in conjunction with local high-dose radiation of the primary tumors was capable of controlling both the primary tumors and distant metastases in unirradiated lungs (1). In other related studies, moderate doses of radiation in the clinical range, when combined with dendritic stimulation, resulted in an “abscopal” response of a distant tumor growing in a nonirradiated site in the same animal (2). Another presentation described how local radiation of tumor, at moderate doses that kill a relatively small fraction of the tumor cells, can actually modulate the phenotype of tumor cells to make them more sensitive to T cell–mediated killing (3, 4). Other data showed that suppression of inhibitory signals by T-regulatory cells could enhance responses of irradiated tumors in mice (5). In addition to these promising results achieved by modulating various aspects of adaptive immunity, results showing effectiveness of stimulation of innate immunity were also presented. Stimulation of innate immunity with foreign molecules, such as CpG oligodeoxynucleotides, which have evolved from earlier studies of immune stimulation with microbial preps, was highly effective when combined with radiation, resulting in significant radiosensitization of the tumors, as well as immunologic memory for prevention of lung colonization after i.v. injection of the tumor cells (6, 7). Related promising results with vaccines, incorporating CpG or muramyl tripeptide (8), for treatment of spontaneous metastatic tumors in dogs are being extended to future trials that incorporate radiation. These research advances in preclinical models were discussed in the context of developing plans for initiating clinical trials with novel vaccines and radiation. Initial results were presented showing that many prostate cancer patients treated with radiation and vaccine exhibited favorable immune system stimulation.

There was a strong feeling among the participants that radiation-enhanced immune response to cancer can be optimized and applied for clinical benefit. CD8+ T-cell responses are most frequently involved in tumor regression, and there have recently been major advances in our detailed knowledge of antigenic epitopes on human tumors, considerable success in fabricating vaccines to boost specific T-cell responses to these epitopes, and development of sensitive assays for reliably measuring these immune responses in humans. Although such modern immunotherapy approaches show promise, they clearly have yet to make a major impact on cancer treatment as a sole therapy and will most likely need to be combined with conventional cytotoxic regimens to show efficacy. It is therefore timely to re-examine how the immune system responds to the liber-
ation and expression of tumor antigens during a course of radiation therapy, how radiation affects immunologically important properties of host and tumor cells and their microenvironment, and how immune responses might be boosted so as to enhance the probability of achieving local control with radiation therapy and counteract systemic micrometastatic disease.

ASSESSING IMMUNE RESPONSES

A significant challenge for human trials of immune modulation is the heterogeneity of clinical responses, which indicates the importance of measuring both the quality and magnitude of the antitumor immune response. It is therefore important for trials of the influence of combined radiation/immune modulation to assay various immune parameters before and frequently during the therapy. This is usually performed with enzyme-linked immunospot and tetramer assays to assess T-lymphocyte responses to specific tumor epitopes, as well as flow cytometric assays for intracellular cytokines that allow simultaneous T-cell subset identification. The distinction between T cells that have a T-helper (Th)1 profile and produce cytokines such as interferon γ and T cells with a Th2 profile that produce cytokines such as interleukin 4 is important because the former are considered to be more involved in cell-mediated immune reactions and tumor rejection, whereas the latter are more involved in antibody formation. Unfortunately, currently there is no “gold standard” assay that monitors changes in immune status after vaccination that necessarily correlates with clinical responses. For this reason, assays of the avidity (functional quality) of T lymphocytes should be carried out whenever possible because these might give more valuable information, even though these assays require large numbers of lymphocytes, usually necessitating leukophoresis of the patient’s blood. Additional useful information about the modulation of the patient’s response could be obtained by analyzing immune cell populations in regional lymph nodes and in both primary and metastatic tumors. This might provide a better understanding of the basis for the apparent discrepancies often seen between the presence of circulating tumor-specific T cells and lack of tumor regression. Furthermore, because T-cell antitumor reactivity is often inhibited by “suppressor cells,” these assays for CD8+ and CD4+ T-cell activity should be supplemented with tests for T-regulatory cells, especially the recently identified CD4+/CD25+/FoxP3+ subset.

Despite the fact that these measurements of the immune functions of patients are not sufficiently robust to serve as surrogates for predicting clinical outcomes, they should be performed in patients receiving radiation therapy. Very little is known about how radiation-induced tumor cell death is handled by the immune system, and information from these assays will provide correlative information that will help to interpret differences in responses among patients and guide the development of this field.

A key area identified as needing more research emphasis is the effect in patients of radiation therapy alone on immune functions. Before embarking on detailed studies of integration of vaccines and co-stimulatory therapy with radiation, we need to understand more about the effects of radiation dose, schedules, and regional treatment fields (that might include lymph nodes) on the immune system. This needs to be assessed with advanced modern immunologic assays.

Also critical for translational research is an understanding of the relevance of the different animal models that are being used. Early-stage experiments are often carried out in murine systems with quite immunogenic tumors. Such systems have the attribute of being more sensitive to immune modulation strategies and can provide proof-of-principle data, as well as guidance for optimizing details of scheduling and integration of immune-based therapy with radiation and chemotherapy. However, results of this research need to be tested on less immunogenic, transplantable rodent tumors that more closely simulate human spontaneous tumors. In addition, companion animals, such as dogs and cats, with certain types of spontaneous cancer provide relevant models of human cancer. Metastatic diseases in dogs, such as osteosarcoma and melanoma, are very similar to those diseases in humans and represent similar heterogeneous tumor populations. Detailed studies of immune modulation are facilitated by standard veterinary practices of anesthetizing for delivery of each dose fraction of radiation therapy, such that biopsy tissue specimens and blood can be readily and serially obtained. Another advantage is that clinical responses can be assessed by human-scale clinical imaging methods, and the time required to assess outcomes is generally considerably less than in humans.

MODIFYING IMMUNE RESPONSE TO TUMORS

Radiation treatment of the tumor can be combined with modulation of the immune system to enhance response of both the primary tumor and of distant metastases, and trials along these lines should be encouraged. In view of the advances in understanding the functioning of the immune system in response to cancers, and of the development of many novel vaccines, it is timely to consider issues related to methods of optimizing the potential of radiation to enhance the immune response to cancers. One relevant observation is the development in the immunized host of cross-priming/sensitization to antigens in addition to the specific antigen(s) in the vaccine. This “antigen cascade” has been seen in analyses of immune responses in both murine models and humans and might be as strong as or even greater than the response to the tumor-associated antigen(s) in the priming vaccine (9). It would be of value to know whether radiation therapy promotes such broadly specific responses. This observation has important implications regarding what should be measured to assess the extent of the immune stimulation in the setting of radiation therapy. It is recommended that, in addition to assaying for responses to the primary sensitizing antigen in the vaccine, tests of other antigens previously demonstrated in the literature to elicit
immune responses in a particular tumor type should be carried out. Obviously, of particular interest would be responses to potential antigens revealed after treatment of the tumor with radiation, although further research would be required to identify these and incorporate them into the measurement of immune sensitization.

Strategies that have been explored for modulation of immunity, in conjunction with radiation therapy, include provision of both antigenic and co-stimulatory signals to enhance adaptive immunity, and stimulation of innate immune receptors for pathogen-associated molecular patterns. Another strategy to overcome host tolerance to the tumor is to inhibit suppressor T cells (T-regulatory cells) with specific antibodies. This has shown clinical efficacy, although it has been associated with undesirable side effects of generalized autoimmunity. Future trials will be aimed at minimizing the autoimmune reactions while maximizing the favorable response.

The route of delivery and the scheduling of the components of the immune modulation protocol require special consideration when combined with radiation therapy. There are data suggesting that intratumoral and s.c. injections might be optimal for some vaccine/co-stimulatory molecule combinations. Beginning the vaccine injections before starting the radiation therapy might also improve the response. There are data that suggest that periodic booster injections should be continued after radiation therapy.

What are the mechanisms by which radiation enhancement of immunity might be achieved? A combination of intratumoral effects of radiation could contribute to the outcomes. The irradiated tumor undergoes a number of changes that might enhance its antigenicity and affect its ability to act as a target for the immune system. A “radiation vaccination” effect might result that would include the enhanced access by dendritic cells to antigenic epitopes from dying cells, and the changes in expression of tumor genes and their coded proteins that contribute to the antigenic milieu. Radiation-induced enhancement of tumor necrosis factor (TNF) and TNF receptor family members, such as fas, or major histocompatibility complex class I, can contribute to rendering the cells more susceptible to immune attack. Post-radiation alterations in the tumor microenvironment and expression of cell adhesion molecules, such as intercellular adhesion molecule 1, might make the tumor more penetrable by immune cells. Recently, direct evidence for radiation-induced enhanced trafficking of antitumor immune effector cells to tumors has been reported. An added effect might result from radiation-induced vascular or lymphatic changes that enhance migration of dendritic cells from the tumor site to the draining lymph nodes. Inhibitory radiation effects on dendritic cells have been reported that might be due to inhibition of proteasome activity and result in a block in antigen processing. On the other hand, in the same study, irradiation was able to stimulate the presentation of exogenous peptides. It is also conceivable that radiation might affect suppressive activity of T-regulatory cells. Old reports that T-suppressor cells are radiosensitive need to be updated with new markers for T-regulatory cells. In all of these situations, radiation dose and timing require investigation, especially with respect to administration of immunomodulatory vaccines. However, the potential rewards are great, and further studies are certainly warranted.

CONCLUSIONS

Continued research on preclinical models is a necessary prerequisite to the implementation of improved methods of combining radiation and immunotherapy to treat human tumors. Clinically, however, the stage is set to expand studies of immune therapy of human cancers to optimize the enhanced responses for primary and metastatic disease that might be produced when immunotherapy is combined with radiation therapy.

REFERENCES