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REVIEW ARTICLE

A perspective on the impact of radiation therapy on the immune rheostat

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ABSTRACT

The advent and success of immune checkpoint inhibitors (ICIs) in cancer treatment has broadened the spectrum of tumours that might be considered "immunogenic" and susceptible to immunotherapeutic (IT) intervention. Not all cancer types are sensitive, and not all patients with any given type respond. Combination treatment of ICIs with an established cytotoxic modality such as radiation therapy (RT) is a logical step towards improvement. For one, RT alone has been shown to be genuinely immunomodulatory and secondly pre-clinical data generally support combined ICI-RT approaches. This new integrated therapy for cancer treatment holds much promise, although there is still a lot to be learned about how best to schedule the treatments, manage the toxicities and determine what biomarkers might predict response, as well as many other issues. This review examines how RT alters the immune rheostat and how it might best be positioned to fully exploit IT.

INTRODUCTION

In the past 2 years, >100 clinical trials have been registered aiming to target negative regulators of T-cell immune activation, known as immune checkpoint inhibitors (ICIs) with local radiation therapy (RT) for cancer treatment (https://clinicaltrials.gov).[1] This burst of enthusiasm for RT-ICI combination therapy has been occasioned by the confluence of several factors. Foremost was the success of the Food and Drug Administration-approved ICIs, ipilimumab (anti-CTLA-4, Yervoy), pembrolizumab (anti-PD-1, Keytruda), nivolumab (anti-PD-1, Opdivo), atezolizumab (anti-PD-L1, Tecentriq) and avelumab (anti-PD-L1, MSB0010718C) as single anti-cancer agents; initial studies in melanoma and renal cancer gave dramatic and durable responses with tolerable toxicity in a significant number of patients.[2] These diseases were chosen because they were considered "immunogenic", largely because they respond to high-dose IL-2. Clinicians were so encouraged that they treated many other cancer types. Some patients with lung, oesophageal, Merkel cell, cervical, gastric, bladder, triple negative breast, colorectal and Hodgkin's cancer responded, leading to the suggestion that these too might be "immunogenic", although in reality only a minority of patients respond to ICIs, while some cancer types seem totally refractory. This was, however, sufficiently enticing to make combinations with an established, locally delivered, highly cytotoxic therapy such as RT, an obvious next step, and encouraged in part by some initial largely anecdotal, but dramatic, successes.[3,4] Other factors driving these trials included the highly competitive market for ICI development, and the growing realization that RT *per se*, even without ICIs, can shift the immune rheostat to influence the elimination of tumours.

Recently, it has become clear that the pathways by which CTLA-4 and PD-1/PD-L1/2 inhibit immune responses are largely distinct,[5] and this might affect the way ICIs interact with RT; something that may also vary with the clinical setting. CTLA-4 is a CD28 homolog that acts early to inhibit immune responses, primarily by affecting the interaction between T cells and antigen-presenting cells (APC) in the lymph nodes, by competing in CD28-mediated co-stimulation and is important for the action of T-regulatory cells (Tregs). PD-1/PD-L1 or PD-1/PD-L2 interactions suppress T cells later on in the effector phase of the response, especially in the periphery. And while anti-CTLA-4 increases T-cell activation primarily by enhancing proliferation and reducing Treg immunosuppression, anti-PD-1/PD-L1 appear to restore activity to "exhausted" T cells. How best to combine ICIs with RT and what to expect in terms of outcomes and toxicities have become the subject of much discussion.[6,7]

INFLAMMATION LINKS RADIATION THERAPY AND IMMUNITY

Radiation oncologists have spent the past century trying to optimize the way to deliver high doses to tumours with acceptable normal tissue toxicity. A seminal discovery in the 1930s that has stood the test of time is that delivering dose in low fractions of about 2 Gy daily over 5–6 weeks spares normal tissues, in particular those that are proliferating more slowly, compared with the tumour.[8] In the early days of RT, the minimal skin erythema dose was an accepted measure to ensure that normal tissue reactions were not excessive. This inflammation, along with the overheating of the low-power X-ray machines of the time, probably promoted the use of dose fractionation, together with experimental evidence that fractionation spared skin testicular reactions in rams being sterilized by irradiation.[9] It became accepted that higher doses of RT are pro-inflammatory, whereas lower doses of ionizing radiation could actually inhibit inflammation. Fraction sizes of around 2 Gy became the convention in the field, with justification for sparing of slowly proliferating normal tissues compared with rapidly proliferating tumours provided by the four R's of radiotherapy—repair, repopulation, redistribution and reoxygenation.[10] Nevertheless, the desire to shorten treatment times continued to drive multiple efforts towards employing higher sized doses for cancer RT. Most often, these resulted in poorer outcomes, although radiobiological modelling was able to direct altered fractionation schemes to moderate success in Phase III clinical trials, validating their use for fraction sizes around 1–3 Gy.[11]

The clinical landscape changed, however, with the introduction of intensity-modulated RT (IMRT) that uses computer-controlled linear accelerators to deliver radiation doses that conform more precisely to the three-dimensional shape of the tumour and to minimize the volume of normal tissues receiving high radiation doses. IMRT has been used to advantage, most notably to give large ablative RT doses to small volumes, or in the treatment of individual or oligometastatic disease sites. Hypofractionation giving doses of 5–10 Gy in 10–5 fractions also draws from this technique and is especially useful to shorten treatment times in situations where conventional (2-Gy) fractionated RT confers little biological advantage because tumour and normal tissue respond to fractionation similarly, or when critical structures can be avoided, or the target volume can be restricted. Although established safety criteria for RT should not be violated when ICIs are added, the evidence suggests that the pro-inflammatory effects of RT kick in only above a certain dose threshold and that hypofractionation with IMRT may be better at generating a state of radiation-induced inflammation that can be exploited immunologically.[12] In other words, isoeffective doses of cytotoxic therapy can be given with a size of dose per fraction chosen to encourage the creation of a "dangerous" microenvironment optimal for engaging the adaptive immune system,[13] which may be of special advantage in the context of concurrent ICI administration with RT. About half the current clinical trials with ICIs plus RT use hypofractionation, and it will be of interest to see if an effect of size of dose per fraction can be discerned.

From a philosophical point of view, evolution did not create processes to directly control tumours, as most arise after the reproductive age, or to cope with high radiation doses. These were unlikely to rank as high evolutionary priorities, unlike the canonical inflammatory processes that combat pathogens, mediate wound healing and maintain immune homeostasis. The normal roles of CTLA-4 and PD-1/PD-L1 are to prevent autoimmunity and control chronic inflammation so as to minimize immune-related normal tissue damage, not to frustrate antitumour IT. The common ground for collaboration between RT and immunotherapy (IT) comes indirectly through inflammation and the links between innate immune mechanisms to adaptive immunity. Inflammation is a prerequisite for maturation of dendritic cells for optimal antigen presentation, T-cell stimulation and the generation of tumour-specific immunity, and doses of radiation that create a "dangerous" inflammatory microenvironment may better assist the development of antitumour immunity.[12] Hard data as to the optimum dose for this are sparse, but production of radiation-induced inflammatory cytokines in tissues peaks around doses of 6–8 Gy, with fractionation extending the response.[14–16] Comparison of fractionation schemes should really be performed at isoeffective levels of cell killing in the presence and absence of an adaptive immune system, something that is rarely carried out, but there is sufficient evidence suggesting that there might be a sweet spot where fractionation is most likely to assist immunity. Hypofractionation with 6–8 Gy appears superior to conventional doses[17] and are more effective than lower or higher doses at generating "abscopal" effects in pre-clinical tumour models, which is when RT delivered to one tumour affects the growth of another at a distant site.[18,19] This suggests that single doses may be less effective than hypofractionated doses at generating immunity, but further confirmation of this in different models is needed. The finding that radiation acts as an immunological adjuvant to increase tumour-specific immune responses has been shown in several tumour models[20,21] but is far from being a universal phenomenon.[22,23] It is worth considering some of the reasons why.

TUMOUR IMMUNOGENICITY, IMMUNOTHERAPY AND RADIATION THERAPY

For the same level of intrinsic tumour radiation sensitivity, tumours that generate effective anti-tumour immunity appear to be more radiosensitive.[17,24] Immunity is therefore a likely cause of radiocurability.[25–27] Indeed, if T-cell depletion increases the dose required to cure tumours (TCD50), it is reasonable to assume that an effective immune response is present. For tumour regression, immunity along the $CD8^+$ T-cell-mediated axis is generally required. From infectious disease studies, we know that this form of immunity is primarily required for the elimination of virus-infected cells. In this respect, it will be fascinating to see the extent to which the known radiosensitivity of HPV+ tumours compared with HPV– tumours has an immunological content. Indeed, there may be fundamental differences in T-cell recognition depending on the nature of their induction, *e.g.* virus-induced *vs* chemically induced tumours. Differences in intratumoural immune cell infiltrates suggest immune involvement,[28] and there are clear genomic differences, although other intrinsic mechanisms have been suggested including differential DNA repair and differences in cell cycle control and in repopulation. It is of course a vast oversimplification to consider the $CD8^+$ T-cell subset as working in

isolation. Anti-tumour responses are multifaceted involving, additionally, $CD4^+$ T-cell helper and regulatory subsets and myeloid cells, not to mention many other cell-associated and soluble molecules, with the vasculature and stromal cells playing major roles. The overall response is an integration of these diverse elements; however, the pre-clinical data clearly ascribes tumour regression to the $CD8^+$ T-cell subset. This raises questions as to the nature of the antigens being recognized, their source and their dependency on the nature of cancer causation.

Tumour regression requires antigen expression and provision of targets, and forced expression of a foreign model antigen, such as ovalbumin, or immune cytokines, such as IL-2, IL-3, IL-4, IL-7 etc.,[14] generates more immunity and makes tumours more radiosensitive. Similarly, the combination of ICIs and RT improves outcomes in pre-clinical studies[29–32] through non-redundant mechanisms.[33] The question as to whether ICI and/or RT can generate immunity against non-immunogenic tumours, however, remains largely unanswered. Although this has been observed in one model after cytokine gene transfection,[34] the extent to which this is generally possible remains uncertain. If anti-tumour immunity is present prior to treatment, removal of immune checkpoints by ICIs or removal of the tumour burden by any means may result in immune activation and regression. On the other hand, there may be little advantage to the use of ICIs if valid regression antigens are not present, but if immunity is generated by RT, for which there is some tenuous clinical evidence,[35] the combination will be more broadly valuable. This steers the discussion towards the nature of tumour-rejection antigens.

Graft rejection and tumour immunology used to be one scientific field before inbred mouse strains were shown to develop tumours that could be successfully transplanted into mice of the same strain but not into mice of a different strain where histocompatibility differences lead to only sporadic tumour take.[36] However, even before these fields diverged, chemically induced tumours were known to be immunologically unique,[37,38] and tumour biopsies could be used as vaccines that would increase the incidence of radiation-induced regression.[39] When syngeneic transplantable tumours became models of choice, it rapidly became clear that chemically or virally induced tumours were much more immunogenic than spontaneous ones. This also raised doubts as to the relevance of chemical-/virus-induced tumours to the human situation which was assumed to be non-immunogenic for the most part.[40] This perspective lasted for decades and changed dramatically only recently with the finding that human tumours vary hugely in the number of mutations they carry and with the notion that higher mutational load goes hand in hand with higher immunogenicity and responsiveness to IT, although the correlation is far from being perfect[41–43] and may not even apply to virus-induced tumours.[44] Tumours that have mutational signatures associated with ultaviolet, chemical carcinogens, age and DNA repair defects, however, can have a high number of tumour mutations and appear able to generate clonal unique antigens that can activate T cells.[45] Because of their immunological component, they can respond to ICI treatment[46] and may be generally more radiosensitive, although many other features have to be taken into account. So, remarkably, the older pre-clinical studies with chemically and virally induced tumours have regained their relevance and can no more be dismissed. An important point needs to be stressed here, namely that the spectrum of mutations in every tumour type is wide, *i.e.* not every melanoma will be highly immunogenic, possibly indicating differences in tumourigenesis, nor will the response to therapy necessarily be predictable. A case in point is that lung cancers from smokers are more immunogenic and ironically respond to ICI therapy better than those from non-smokers.[43] However, in a RT setting without ICIs, any benefit to patients from tumour immunity is likely offset by smoking causing hypoxia, which increases radioresistancy, let alone the increased risks of second cancers and cardiovascular events.[47]

The exact nature of tumour-associated antigens that can actually direct T-cell-mediated tumour regression is still elusive. Clearly, they form a small minority of conformally altered molecules expressed by tumours. Chemical- or ultraviolet-induced human and animal tumours may have unique non-cross-reacting regression antigens, but little is known about what the immune system sees in virus-induced tumours. However, the findings clearly indicate that some human tumours express molecules that autologous $CD8^+$ T cells can recognize and by which they can be activated.[48,49] Currently, the expression of very restricted T-cell repertoires in tumour-infiltrating T cells (TIL) is taken as evidence of this and of an ongoing clonal selection process, as was suggested by early studies comparing tumour infiltrating, blood and lymph node lymphocytes.[48] In general, the presence of pre-existing tumour immunity in patients when they come in for treatment with ICIs and/or RT is a good prognostic sign, but the real test for broad therapeutic applicability is whether immunity can be generated *de novo*. RT may help this process as occasional conversions to responsiveness have been detected following RT,[35] but further studies are needed. Obviously, patients with advanced cancer are often immunologically inert as a result of their chronic condition and it will be interesting to see if they can be immunologically reactivated by ICIs and/or RT. It should be noted that tumour-immune regression as an end point is very different from that of regression during chemotherapy or RT. The timing of response will be different, and it may take a considerable amount of time, but it is generally associated with durable complete tumour elimination with limited or no normal tissue toxicity.

RADIATION-INDUCED DANGER, INFLAMMATION AND IMMUNITY

To make a successful immune response, such as against a tumour, a pro-inflammatory, oxidatively stressed "dangerous" microenvironment is created through the release of damage-associated molecular pattern molecules, such as the "danger" signals ATP and HMGB1. This promotes an immunological ballet between different immune cells. It is orchestrated by the ascendency of a long list of pro-inflammatory cytokines and chemokines including IFN-γ, GM-CSF, TNF-α, IL-1, IL-2, IL-3 and IL-4 and involves local draining lymph nodes and even distant sites where the orchestra may be playing different tunes. T-helper cells play a major controlling role by directing the response. In this way, the immune rheostat is set at a point that

allows maturation of dendritic cells (DCs) for antigen presentation, up-regulation of major histocompatibility complex (MHC) Class I and II to facilitate immune recognition while other cell adhesion systems feed forward to promote vascular activation and further immune cell infiltration. This disturbance of the normal homeostatic equilibrium is required to generate immunity but with restraint systems kicking in an attempt to re-exert control. These include induction of pathways such as PD-L1 and PD-1, T regulatory and myeloid suppressor cells and the anti-inflammatory cytokines IL-10 and TGF-β that aim to counterbalance the pro-inflammatory, pro-oxidant, immune-activating processes. This yin–yang of dueling, mutually antagonistic forces is inherent in the immune system and has implications for cancer IT and RT, not the least being that as the rheostat moves towards restoring control and a general wound healing setting in which tumour growth and metastasis can be accelerated, *e.g.* by myeloid suppressor cell involvement.

Ionizing radiation is able to mimic most of the microenvironmental "danger" responses and can act as a bona fide "danger" signal. Not only can it assist in the generation of an immune state but also turn tumour cells into better targets by increasing expression of MHC Class I and death family receptors. Additional reinforcing signals may come from calreticulin and phosphatidyl serine expression on tumour cells during radiation-induced "immunogenic" cell death,[50] which may be a direct result of the oxidative stress conditions. Cells dying by radiation-induced apoptosis do not do so "silently", which is important for the generation of a permissive environment and antigen recognition. However, it must be stressed that although RT may appear to drive immunogenicity, evidence that regression antigens are generated *de novo* by RT is lacking, unlike the case with mutagenic chemicals.[51] Therefore, it seems that RT, similar to ICIs, is more likely to rely on pre-existing antigens to direct immune activation. Another big plus of RT is that it can significantly enhance the ingress of immune cells into the tumour.[20] This is likely due to radiation-induced inflammation increasing the vascular flow and tumour cell death decreasing the interstitial fluid pressure, as well as activating cascades of chemokines, cytokines and vascular cell adhesion molecules that promote infiltration. It should be noted that RT may also damage the intratumoural vasculature, especially at higher doses,[52] potentially hindering immune infiltration, which suggests that IT may be best given prior to RT for which there is some evidence.[22] ICI treatment, such as RT, increases the T-cell infiltrate into tumours,[53,54] and the combination therapy might therefore generate a superior source of tumour-infiltrating T cells for adoptive transfer, especially if ICI-RT treatment was to be given prior to tumour resection.

As might be expected, RT can mature DCs and enhance their ability to cross-present immunodominant MHC-Class I peptides so as to generate superior immunity and tumour rejection.[55,56] Cross-priming and cross-presentation is how the immune system detects and responds to viral or mutational antigens that occur in tumours or normal cells and that have to be processed and presented by professional APC. The APC acquire proteins from other tissue cells through endocytic mechanisms and process them either by proteasomal degradation or endosomal proteases. Either way, they get loaded onto the MHC molecules. The dominance of the processing pathway that is used probably depends on the nature of the antigen, the nature of the microenvironment and the state of the APC, with both tolerance and immunity as possible outcomes. Both radiation and proteasome inhibition enhance DC cross-presentation of loaded peptides and their ability to generate immunity.[55] By contrast, irradiation of DCs prior to delivery of whole antigen by adenoviral vectors, blocks their ability to generate immunity.[53] This suggests that RT can determine the choice of antigen-processing pathways, which may be a natural rheostat by which DCs control immunity/tolerance induction in response to the changing microenvironment and "danger" signalling. A case in point is the ability of chloroquine—an endocytic compartment inhibitor—to modulate radiation-induced tumour immunity presumably by altering antigen degradation.[57] Clearly, the effects of RT on the processing and presentation of antigens are very complex, and it is therefore no surprise that RT can do both, namely enhance immunity as well as drive control mechanisms, including PD-L1/PD-1 and regulatory T cells.

IMMUNE SUPPRESSION, IMMUNOTHERAPY AND RADIATION THERAPY

Immune responses occur in specialized lymphoid organs and at localized sites to prevent severe morbidity or mortality that would arise from systemic immune activation. Control mechanisms, such as CTLA-4, PD-1, Tregs and myeloid suppressor cells have evolved to restrain and localize normal tissue damage following inflammation and immune recognition. One consequence of this trade-off is that the suppressor mechanisms allow even highly immunogenic tumours to grow, as has been evident from experimental animal models for decades. Both ICIs and RT shift this immune rheostat. The art will be to optimally combine these so that they synergize and not negate one another, and to do so without increasing systemic and local toxicity beyond acceptable limits. Biomarkers are urgently needed to assess the position of the immune rheostat in patients as they begin therapy.

The effects of RT on the rheostat determining suppression *vs* immunity are complex with many moving parts. At the simplest level, and known for many decades, RT decreases the number of peripheral lymphocytes, even when given locally.[58–60] In humans, but not mice, this decrease can last for many years, with most subsets affected, and with a decreased T helper : suppressor ratio. This is thought to be due to lymphocytes in the blood being killed as they pass through the radiation field. Of note, the radiosensitivity of lymphocytes is so high that conventional, low dose per fractions are as potent as high dose per fraction in this regard, *i.e.* making volume the main determining factor while the issue of fraction size becomes trivial.[61] The effect of this on anti-tumour responsiveness is not clear but, if depletion is extensive, increased morbidity and infection are seen. Interestingly, ICIs may mitigate this effect, suggesting a limiting step during lymphocyte recovery that is otherwise in place.

In many cases, RT-like surgery can remove large immunosuppressive tumour burdens or at least cause growth arrest. Since

tumour regression is likely to be at some stage a numerical game between the immune and tumour forces, RT may simply tip the balance in favour of the host. Removal of tumour burden may also allow a pre-existing state of anti-tumour immunity to emerge or, alternatively, a state of tumour immunity to be generated. It is hard to distinguish between these possibilities, but they are obviously very different processes. The complexity of the tumour–host relationship in these situations is often overlooked. For instance, there is evidence that small numbers of highly immunogenic tumour cells can "sneak through" the host defenses,[62,63] adjusting the rheostat early on to suppression rather than immunity.[64] Ironically, the size of tumour inocula most often used in experimental studies tends to be also optimal for the generation of immunity.[64] As immunogenic tumours grow, immunity is (and has to be) counteracted by the generation of T regulatory/suppressor cells followed by the development of a non-specific state of immunological inertia probably associated with activation of the myeloid system. It is hard to predict the effect of tumour removal in a host where immunity is suppressed.

Local or systemic RT can increase the numbers and activities of T regulatory cells even in the absence of tumour.[65] This is in part due to their radioresistancy relative to most lymphocytes so that even 2 Gy can increase their representation, but they are also generated just in response to radiation-induced tissue damage. RT can also induce PD-L1 expression on tumour cells and suppress anti-tumour immunity.[66] Yet, even in the presence of suppression, concomitant immunity can coexist and, even though it may not be obvious, it may be effective, *e.g.* in controlling possible metastatic spread by immunogenic tumours.[24] Many other "immune escape" mechanisms have been postulated to explain the growth of tumours in the face of immune aggression that are the subject of excellent reviews and will not be reiterated here.[67,68]

The concept that immunity capable of tumour regression can exist in at least some cancer patients while being held in check by suppressor mechanisms is critical for the consideration of combined ICI + RT treatments, remembering that some patients may display anti-cancer responses that are not CD8-mediated and are therefore less able to mediate tumour cure, even though they may impact tumour growth. Under any circumstances, decreasing the tumour burden should be a primary therapeutic aim and RT should therefore not be compromised for ICI treatment. In further support of this approach, many publications suggest that non-curative RT and surgery tend to promote metastasis development, which is not the case for curative treatments.[69] By the same token, to rely on immune "abscopal" action to eliminate established tumours, may be overly optimistic given the rarity of such events with RT alone[70] and the general presence of the suppressive state. It should also be noted that tumour shrinkage at a site distant from that irradiated is not necessarily an evidence of generalized immune activation, and involvement as other mechanisms, such as interference with angiogenesis, are conceivable.[71] Until the abscopal effect is shown to be a common effect of ICI plus RT, in our opinion oligometastatic disease sites should not be left untreated, even in the context of ICI treatment. Perhaps a more realistic aim is to engage the immune system to assist in the permanent elimination of established and micrometastatic tumours and to develop lasting immunological memory to minimize recurrence.

A broad generalization is that at the other end of the immunogenic spectrum, there will be many tumours that have a low mutational load, no evidence of virus, ultraviolet, chemical induction or DNA repair-deficiency and respond poorly to ICIs. Animal studies suggest that the myeloid axis may be of more relevance for such tumours, although this almost bound to be an oversimplification given the complex interplay within the immune system. The role of myeloid cells in tumourigenesis, metastatic potential and T-cell control is not within the remit of this review but some points have to be made. Tumours have been referred to as "wounds that do not heal",[72] which implies involvement of myeloid cells in attempted healing responses. The known plasticity and diversity within and between subsets of the myeloid system,[73] the difficulty in eliminating them by genetic manipulations or drug treatments, and in reliably isolating them from tissue sites without altering their functional expression makes it hard to evaluate the role of these cells in cancer development and therapy. Myeloid cells, especially macrophages, are major cellular components in tumours where they express much heterogeneity.[74,75] The relationship between chronic inflammation and cancer development implicates myeloid cells in carcinogenesis, as well as on tumour growth promotion. RT activates myelogenesis, whether bone marrow is in the field or not,[76] and can stimulate macrophages to further promote tumour growth.[77,78] On the other hand, RT also functionally alters macrophages to prime them for production of pro-inflammatory cytokines, displaying the yin–yang relationships within the myeloid system.[14,15] There are several therapeutic approaches that block radiation-induced myeloid cell mobilization and augment the effects of RT.[76,79] The extent to which these approaches result in effective antitumour immunity leading to tumour regression is not clear; blocked angiogenesis and loss of growth stimulation may contribute more than T-cell activation to the therapeutic outcome.

PD-L1 is expressed by many cells upon activation and in response to many signals. The interferons are especially effective, but RT also increases expression.[80] This is a reflection of the role of these molecules in feedback control of inflammation and immunity, which takes the PD-1/PD-L1/2 axis far outside the realm of controlling just T-cell-mediated responses. PD-L1 upregulation on myeloid cells, along with the sheer number and exquisite mobility of macrophages, makes them possible important targets for ICI therapy. Certainly, the integration of CSF1R inhibitors, or other inhibitors of myeloid cell activation, into ICI/RT regimes is of huge interest. However, if tumours with a strong myeloid component prove to be a separate category from those that predominantly recruit lymphoid cells, such combinations may not be very effective indeed.

TOXICITIES OF IMMUNE CHECKPOINT INHIBITOR/ RADIATION THERAPY

Ultimately, the art of combining ICIs with RT may be to select patients in whom the immune rheostat is set to a position where

tumour-specific immunity can be liberated without excessive adverse side effects. ICIs with RT may not result in *de novo* generation of tumour regression antigens but should reset the immune rheostat towards increasing immunity to existing antigens. Both, however, will have local and systemic consequences that have their basis in autoimmunity and inflammation and will lead to tissue damage.

The toxicities associated with conventional RT are generally well known, as are the limits of hypofractionated RT. The toxicities associated with ICI have also been documented and are often referred to as "immune-related adverse events" (irAEs) that are overwhelmingly related to breaking tolerance and the manifestation of autoimmune reactions,[81] with slight variations depending on the nature of the ICI. Low-grade irAE is seen in over half the patients receiving ICIs, but serious adverse reactions are relatively rare with $<1\%$ mortality.[81] Ipilimumab has been reported to have about twice as many grade ≥ 3 irAEs than pembrolizumab.[82] Combination of different ICIs increase toxicity and the addition of RT might be expected to drive it further, if for no other reason than it is pro-inflammatory. Any organ system may be affected, but lesions involving the gut, skin, liver, thyroid and lung are most common. These are generally easy to mitigate with steroids, although cardiovascular toxicity from autoimmune myocarditis, cardiomyopathy, heart failure, cardiac fibrosis and cardiac arrest, while rare, may lead to significant morbidity and mortality[83] in line with the finding that PD-1 in animals confers protection against inflammation and heart damage.[84]

CONCLUSION

The enthusiasm with which ICI therapies have been integrated into RT schedules is remarkable and the results on the ongoing >100 clinical trials are awaited with interest. While biologics have been introduced into radiation oncology clinics before, none have come close to penetrating a specialty where most of the clinical trials have been driven largely by advances in physics. Some clarity is emerging on how best to combine ICIs with RT. One thing ICIs and RT have in common is that they can alter immune rheostats so as to enhance inflammation and the level of anti-tumour immunity, but by different pathways. Tumour immunogenicity may be the major factor deciding if specific antitumour immunity is engaged that can enhance radiation-induced regression and long-term cure, and since ICIs do not seem to increase tumour immunogenicity, the potential of RT to do so is likely to be important in determining the utility of the combinations. Inhibitors of myeloid suppressor cells would be expected to be similarly limited to more immunogenic tumours. Pre-clinical data suggest that hypofractionated RT may be superior to conventional or single-dose RT in enhancing tumour immunity, although this requires further clinical confirmation. Because both ICIs and RT are generally pro-inflammatory, it seems very possible that combinations will uncover some unexpectedly increased late inflammatory or autoimmune toxicities. It is therefore important that the established principles of radiobiology as they relate to therapy are not compromised and that precision RT is used to minimize high doses to normal tissues. However, with care, IT promises much in being able to augment and extend the reach of RT to cancer situations that are currently treated only with a low chance of success.

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