CHAPTER 2

BASIC RADIOBIOLOGY

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2.1. INTRODUCTION

Radiobiology is the study (both qualitative and quantitative) of the actions of ionizing radiations on living matter. Since radiation has the ability to cause changes in cells which may later cause them to become malignant, or bring about other detrimental functional changes in irradiated tissues and organs, consideration of the associated radiobiology is important in all diagnostic applications of radiation. Additionally, since radiation can lead directly to cell death, consideration of the radiobiological aspects of cell killing is essential in all types of radiation therapy.

2.2. RADIATION EFFECTS AND TIMESCALES

At the microscopic level, incident rays or particles may interact with orbital electrons within the cellular atoms and molecules to cause excitation or ionization. Excitation involves raising a bound electron to a higher energy state, but without the electron having sufficient energy to leave the host atom. With ionization, the electron receives sufficient energy to be ejected from its orbit and to leave the host atom. Ionizing radiations (of which there are several types) are, thus, defined through their ability to induce this electron ejection process, and

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the irradiation of cellular material with such radiation gives rise to the production of a flux of energetic secondary particles (electrons). These secondary particles, energetic and unbound, are capable of migrating away from the site of their production and, through a series of interactions with other atoms and molecules, give up their energy to the surrounding medium as they do so.

This energy absorption process gives rise to radicals and other chemical species and it is the ensuing chemical interactions involving these which are the true causatives of radiation damage. Although the chemical changes may appear to operate over a short timescale (~10^{-5} s), this period is nonetheless a factor of ~10^{18} longer than the time taken for the original particle to traverse the cell nucleus. Thus, on the microscopic scale, there is a relatively long period during which chemical damage is inflicted (Table 2.1).

It is important to note that, irrespective of the nature of the primary radiation (which may be composed of particles and/or electromagnetic waves), the mechanism by which energy is transferred from the primary radiation beam to biological targets is always via the secondary electrons which are produced. The initial ionization events (which occur near-instantaneously at the microscopic level) are the precursors to a chain of subsequent events which may eventually lead to the clinical (macroscopic) manifestation of radiation damage.

Expression of cell death in individual lethally damaged cells occurs later, usually at the point at which the cell next attempts to enter mitosis. Gross (macroscopic and clinically observable) radiation effects are a result of the wholesale functional impairment that follows from lethal damage being inflicted to large numbers of cells or critical substructures. The timescale of the whole process may extend to months or years. Thus, in clinical studies, any deleterious health effects associated with a radiation procedure may not be seen until long after the diagnostic test or treatment has been completed (Table 2.1).

TABLE 2.1. THE TIMESCALES OF RADIATION EFFECTS

Action	Approximate timescale
Initial ionizing event	10^{-18} s
Transit of secondary electrons	10^{-15} s
Production of ion radicals	10^{-10} s
Production of free radicals	10^{-9} s
Chemical changes	10^{-5} s
Individual cell death	Hours–months
Gross biological effects	Hours–years

2.3. BIOLOGICAL PROPERTIES OF IONIZING RADIATION

2.3.1. Types of ionizing radiation

In nuclear medicine, there are four types of radiation which play a relevant role in tumour and normal tissue effects: gamma (γ) radiation, beta (β) radiation, alpha (α) particles and Auger electrons.

2.3.1.1. Gamma radiation

Gamma radiation is an electromagnetic radiation of high energy (usually above 25 keV) and is produced by subatomic particle interactions. Electromagnetic radiation is often considered to be made up of a stream of wave-like particle bundles (photons) which move at the speed of light and whose interaction properties are governed mainly by their associated wavelength. Although the collective ionization behaviour of large numbers of photons can be predicted with great accuracy, individual photon interactions occur at random and, in passing through any type of matter, a photon may interact one or more times, or never. In each interaction (which will normally involve a photoelectric event, a Compton event or a pair production event), secondary particles are produced, usually electrons (which are directly ionizing) or another photon of reduced energy which itself can undergo further interactions. The electrons undergo many ionizing events relatively close to the site of their creation and, therefore, contribute mostly to the locally absorbed dose. Any secondary photons which may be created carry energy further away from the initial interaction site and, following subsequent electron-producing interactions, are responsible for the dose deposition occurring at sites which are more distant from the original interaction.

2.3.1.2. Beta radiation

Beta radiation is electrons emitted as a consequence of β radionuclide decay. A β decay process can occur whenever there is a relative excess of neutrons (β^-) or protons (β^+). One of the excess neutrons is converted into a proton, with the subsequent excess energy being released and shared between an emitted electron and an anti-neutrino. Many radionuclides exhibit β decay and, in all cases, the emitted particle follows a spectrum of possible energies rather than being emitted with a fixed, discrete energy. In general, the average β energy is around one third of the maximum energy. Most β emitting radionuclides also emit γ photons as a consequence of the initial β decay, leaving the daughter nucleus in an excited, metastable state. Since β particles are electrons, once ejected from the host atom,

they behave exactly as do the electrons created following the passage of a γ ray, giving up their energy (usually of the order of several hundred kiloelectronvolts) to other atoms and molecules through a series of collisions.

For radionuclides which emit both β particles and γ photons, it is usually the particulate radiation which delivers the greatest fraction of the radiation dose to the organ which has taken up the activity. For example, about 90% of the dose delivered to the thyroid gland by ^{131}I arises from the β component. On the other hand, the γ emissions contribute more significantly to the overall whole body dose.

2.3.1.3. Alpha particles

Alpha radiation is emitted when heavy, unstable nuclides undergo decay. Alpha particles consist of a helium nucleus (two protons combined with two neutrons) emitted in the process of nuclear decay. The α particles possess approximately 7000 times the mass of a β particle and twice the electronic charge, and give up their energy over a very short range (<100 μm). Alpha particles usually possess energies in the megaelectronvolt range, and because they lose this energy in such a short range are biologically very efficacious, i.e. they possess a high linear energy transfer (LET; see Section 2.6.3) and are associated with high relative biological effectiveness (RBE; see Section 2.6.4).

2.3.1.4. Auger electrons

Radionuclides which decay by electron capture or internal conversion leave the atom in a highly excited state with a vacancy in one of the inner shell electron orbitals. This vacancy is rapidly filled by either a fluorescent transition (characteristic X ray) or non-radiative (Auger) transition, in which the energy gained by the electron transition to the deeper orbital is used to eject another electron from the same atom. Auger electrons are very short range, low energy particles that are often emitted in cascades, a consequence of the inner shell atomic vacancy that traverses up through the atom to the outermost orbital, ejecting additional electrons at each step. This cluster of very low energy electrons can produce ionization densities comparable to those produced by an α particle track. Thus, radionuclides which decay by electron capture and/or internal conversion can exhibit high LET-like behaviour close (within 2 nm) to the site of the decay.

2.4. MOLECULAR EFFECTS OF RADIATION AND THEIR MODIFIERS

Radiation induced damage to biological targets may result from direct or indirect action of radiation (Fig. 2.1):

— Direct action involves ionization or excitation (via Coulomb interactions) of the atoms in the biological target. This gives rise to a chain of events which eventually leads to the observable (macroscopic) damage. In normally oxygenated mammalian cells, the direct effect accounts for about one third of the damage for low LET radiations such as electrons and photons.
— Indirect action involves radiation effects on atoms or molecules which are not constituent parts of the biological target. Since cells exist in a rich aqueous environment, the majority of indirect actions involve the ionization or excitation of water molecules. The free radicals subsequently created may then migrate and damage the adjacent biological targets. Indirect action is the main cause of radiation damage and, in normoxic cells, accounts for about two thirds of the damage.

Indirect action is predominant with low LET radiation, e.g. X and γ rays, while direct action is predominant with high LET radiation, e.g. α particles and neutrons.

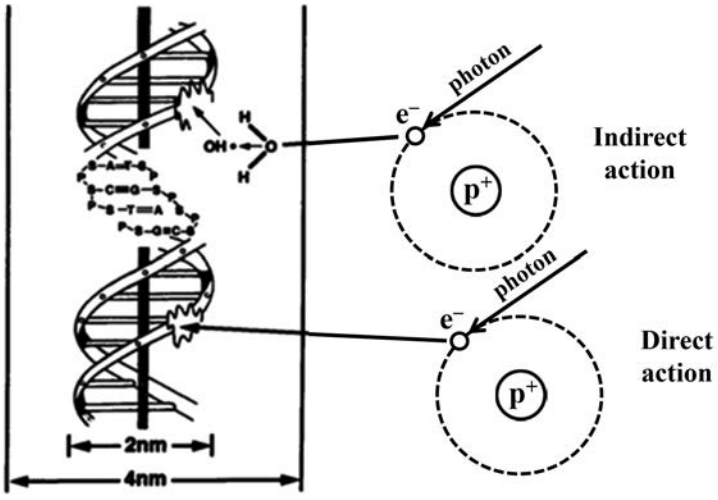

FIG. 2.1. Illustration of the difference between direct and indirect damage to cellular DNA.

2.4.1. Role of oxygen

Radiation effects may be influenced by several factors, especially the presence or absence of oxygen. The free radicals (denoted by a dot placed to the right of the atomic symbol) produced as a result of direct or indirect effects are very reactive and seek to interact with other molecules which can share or donate electrons. Molecular oxygen (O_2) has two unpaired electrons and readily reacts with free radicals, causing an increased likelihood that DNA (deoxyribonucleic acid) will be damaged by the indirect process. Important reactions via which oxygen can increase biological damage are:

$$R^{\bullet} + O_2 \rightarrow RO_2^{\bullet} \ (\text{highly toxic})$$
$$H^{\bullet} + O_2 \rightarrow HO_2^{\bullet}$$
$$HO_2^{\bullet} + HO_2^{\bullet} = H_2O_2 \ (\text{highly toxic}) + O_2$$

where R represents an organic molecule.

The oxygen enhancement ratio (OER) is given by the dose in hypoxia (total absence of oxygen) divided by the dose in air required to achieve an equivalent biological effect. For low LET radiation, such as γ rays, the OER has a value of ~3. For high LET radiation, such as α particles, the OER decreases to almost 1.0.

2.4.2. Bystander effects

Bystander effects occur when a cell which has not been traversed by a charged particle is damaged as a result of radiation interactions occurring in neighbouring cells. The discovery of the bystander effect poses a challenge to the traditional view that all radiation damage stems from direct interactions of charged particles with critical cellular targets. For this reason, it still remains controversial in radiobiology. A possible explanation is that irradiated cells may send out a stress signal to nearby cells, which may elicit a response, e.g. the initiation of apoptosis, in those cells. The overall relevance of the bystander effect is presently difficult to gauge. It is probably most significant in radiation protection considerations involving low doses since it amplifies the overall radiation effect in situations where not all of the cells in a tissue are subjected to particle transversal, i.e. the overall radiation risk to that tissue is higher than would be expected from consideration of the gross response exhibited by those cells which have been directly traversed by charged particles.

2.5. DNA DAMAGE AND REPAIR

2.5.1. DNA damage

DNA damage is the primary cause of cell death caused by radiation. Radiation exposure produces a wide range of lesions in DNA such as single strand breaks (SSBs), double strand breaks (DSBs), base damage, protein–DNA cross-links and protein–protein cross-links (see Fig. 2.1). The number of DNA lesions generated by irradiation is large, but there are a number of mechanisms for DNA repair. As a result, the percentage of lesions causing cell death is very small. The numbers of lesions induced in the DNA of a cell by a dose of 1–2 Gy are approximately: base damages: >1000; SSBs: ~1000; DSBs: ~40. DSBs play a critical role in cell killing, carcinogenesis and hereditary effects. There are experimental data showing that the initially produced DSBs correlate with radiosensitivity and survival at lower dose, and that unrepaired or misrepaired DSBs also correlate with survival after higher doses. Furthermore, there is experimental evidence for a causal link between the generation of DSBs and the induction of chromosomal translocations with carcinogenic potential.

2.5.2. DNA repair

DNA repair mechanisms are important for the recovery of cells from radiation and other damaging agents. There are multiple enzymatic mechanisms for detecting and repairing radiation induced DNA damage. DNA repair mechanisms, such as base excision repair, mismatch repair and nucleotide excision repair, respond to damage such as base oxidation, alkylation and strand intercalation. Excision repair consists of cleavage of the damaged DNA strand by enzymes that cleave the polynucleotide chain on either side of the damage, and enzymes which cleave the end of a polynucleotide chain allowing removal of a short segment containing the damaged region. DNA polymerase can then fill in the resulting gap using the opposite undamaged strand as a template. For DSBs, there are two primary repair pathways, non-homologous end joining (NHEJ) and homologous recombination. NHEJ repair operates on blunt ended DNA fragments. This process involves the repair proteins recognizing lesion termini, cleaning up the broken ends of the DNA molecule, and the final ligation of the broken ends. DSB repair by homologous recombination utilizes sequence homology with an undamaged copy of the broken region and, hence, can only operate in late S/G2-phases of the cell cycle. Undamaged DNA from both strands is used as a template to repair the damage. In contrast to NHEJ, the repair process of homologous recombination is error-free. Repair by NHEJ operates throughout the cell cycle but dominates in G1/S-phases. The process is error-prone because it

does not rely on sequence homology. Unrepaired or misrepaired damage to DNA will lead to mutations and/or chromosome damage in the exposed cell. Mutations might lead to cancer or hereditary effects (when germ cells are exposed), whereas severe chromosome damage often leads to cell death.

2.6. CELLULAR EFFECTS OF RADIATION

2.6.1. Concept of cell death

Radiation doses of the order of several grays may lead to cell loss. Cells are generally regarded as having been 'killed' by radiation if they have lost reproductive integrity, even if they have physically survived. Loss of reproductive integrity can occur by apoptosis, necrosis, mitotic catastrophe or by induced senescence. Although all but the last of these mechanisms ultimately results in physical loss of the cell, this may take a significant time to occur.

Apoptosis or programmed cell death can occur naturally or result from insult to the cell environment. Apoptosis occurs in particular cell types after low doses of irradiation, e.g. lymphocytes, serous salivary gland cells, and certain cells in the stem cell zone in testis and intestinal crypts.

Necrosis is a form of cell death associated with loss of cellular membrane activity. Cellular necrosis generally occurs after high radiation doses.

Reproductive cell death is a result of mitotic catastrophe (cells attempt to divide without proper repair of DNA damage) which can occur in the first few cell divisions after irradiation, and it occurs with increasing frequency after increasing doses.

Ionizing radiation may also lead to senescence. Senescent cells are metabolically active but have lost the ability to divide.

2.6.2. Cell survival curves

A quantitative understanding of many aspects of biological responses to radiation may be made by consideration of the behaviour of the underlying cell survival (dose response) characteristics. Although the practical determination of cell survival curves is potentially fraught with experimental and interpretational difficulties and is best performed by persons who are experts in such procedures, an appreciation of the structure and meaning of such curves, even in a purely schematic context, can be very helpful in understanding the role played by the various factors which influence radiation response.

Figure 2.2 shows the typical shape of a cell survival curve for mammalian tissue. Physical radiation dose is plotted on the linear horizontal axis while

fractional cell survival is plotted on the logarithmic vertical axis. Each of the individual points on the graph represents the fractional survival of cells resulting from delivery of single acute doses of the specified radiation, which in this case is assumed to be γ radiation. (In the context of the subject, an acute dose of radiation may be taken to mean one which is delivered at high dose rate, i.e. the radiation delivery is near instantaneous.) Mammalian cell survival curves plotted in this way are associated with two main characteristics: a finite initial slope (at zero dose) and a gradually increasing slope as dose is increased.

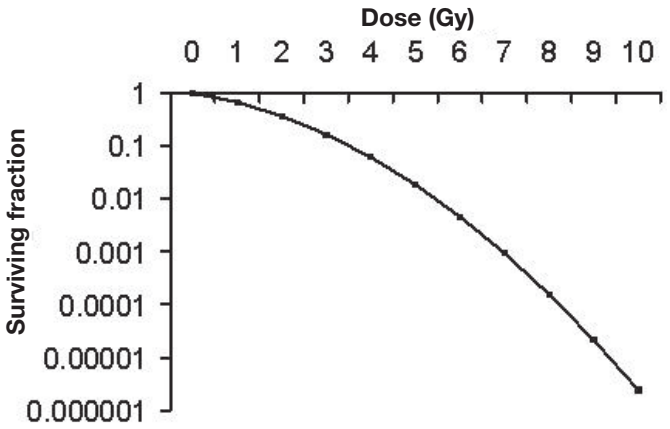

FIG. 2.2. A radiation cell survival curve plots the fraction of plated cells retaining colony forming ability (cell surviving fraction) versus radiation absorbed dose.

2.6.3. Dose deposition characteristics: linear energy transfer

As noted above, the energy transfer to the absorbing medium (whether that be animate or inanimate material) is via secondary electrons created by the passage of the primary ionizing particle or ray. LET is a measure of the linear rate at which radiation is absorbed in the absorbing medium by the secondary particles and is defined by the International Commission on Radiation Units and Measurements (ICRU) as being the quotient dE/dl, where dE is the average energy locally imparted to the medium by a charged particle of specified energy in traversing a distance dl. The unit usually employed for LET is kiloelectronvolt per micrometre and some representative values are listed in Table 2.2.

TABLE 2.2. THE LINEAR ENERGY TRANSFER OF DIFFERENT RADIATIONS

Radiation type	Linear energy transfer (keV/μm)
^{60}Co γ rays	0.2
250 kVp X rays	2.0
10 MeV protons	4.7
2.5 MeV α particles	166
1 MeV electrons	0.25
10 keV electrons	2.3
1 keV electrons	12.3

For radiobiological studies in particular, the concept of LET is problematic since it relates to an average linear rate of energy deposition but, at the microscopic level (i.e. at dimensions comparable with the critical cellular targets), the energy deposited per unit length along different parts of a single track may vary dramatically. In particular, as charged particles lose energy in their passage through a medium via the result of collision and ionizing processes, the LET rises steeply to its highest value towards the very end of their range. The change in LET value along the track length is one reason why average LET values correlate poorly with observed (i.e. macroscopic) biological effects. For these reasons, the directly measured RBE is of much greater use as an indicator of the differing biological efficacies of various radiation types.

2.6.4. Determination of relative biological effectiveness

For a given biological end point, the RBE of the high LET radiation is defined as the ratio of the isoeffective doses for the reference (low LET) and the high LET radiation (Fig. 2.3). The reference radiation is usually ^{60}Co γ rays or high energy (250 kVp) X rays.

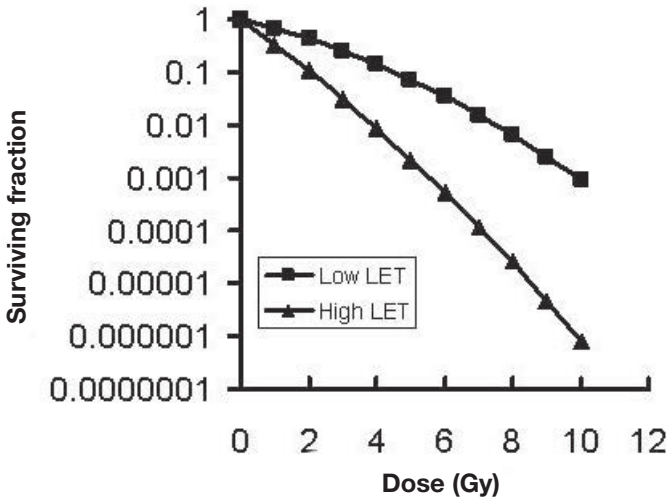

FIG. 2.3. The relative biological effectiveness of a radiation is defined as the ratio of the dose required to produce the same reduction in cell survival as a reference low linear energy transfer (LET) radiation.

If the respective low and high LET isoeffective doses are d_L and d_H, then:

$$\mathrm{RBE} = \frac{d_L}{d_H} \tag{2.1}$$

If the basic cell survival curves are described in terms of the linear–quadratic (LQ) model, then the surviving fraction S as a function of acute doses at low and high LET is respectively given as:

$$S_L = \exp\left(-\alpha_L d_L - \beta_L d_L^2\right) \tag{2.2}$$

$$S_H = \exp\left(-\alpha_H d_H - \beta_H d_H^2\right) \tag{2.3}$$

where the suffixes L and H again respectively refer to the low and high LET instances.

Figure 2.4 shows an example of how the RBEs determined at any particular end point (cell surviving fraction) vary with changing dose for a given radiation fraction size for a low LET radiation. The maximum RBE (RBE_{max}) occurs at zero dose and, in terms of microdosimetric theory, corresponds to the ratio

between the respective high and low LET linear radiosensitivity constants, α_H and α_L, i.e.:

$$\mathrm{RBE}_{\max} = \frac{\alpha_H}{\alpha_L} \tag{2.4}$$

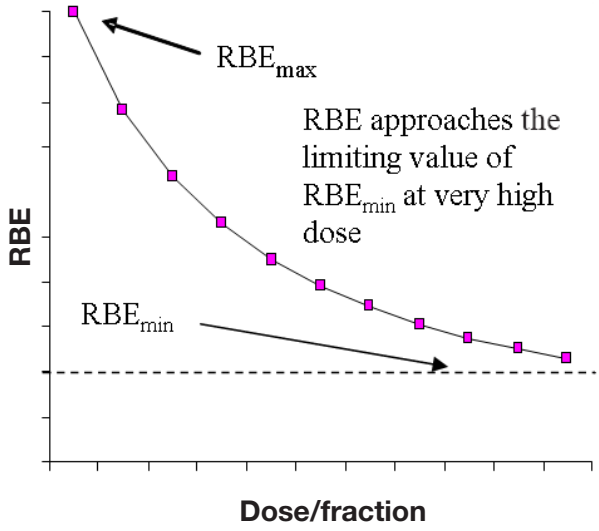

FIG. 2.4. Relative biological effectiveness (RBE) as a function of the radiation dose per fraction.

If the quadratic radiosensitivity coefficients (β_H and β_L) are unchanged with changing LET (i.e. $\beta_H = \beta_L$), then, at high doses, the RBE tends to unity. However, this constancy of β, assumed by the theory of Kellerer and Rossi, has been challenged and, if β does change with LET, then RBE will tend asymptotically to an alternative minimum value ($\mathrm{RBE}_{\min}$) given by:

$$\mathrm{RBE}_{\min} = \sqrt{\frac{\beta_H}{\beta_L}} \tag{2.5}$$

and the 'working' RBE at any given dose per fraction is given as:

$$\mathrm{RBE} = \frac{(\alpha/\beta)_L \mathrm{RBE}_{\max} + \sqrt{(\alpha/\beta)_L^2 \mathrm{RBE}_{\max}^2 + 4d_L \mathrm{RBE}_{\min}^2 \left[(\alpha/\beta)_L + d_L\right]}}{2\left[(\alpha/\beta)_L + d_L\right]} \tag{2.6}$$

when expressed in terms of the low LET dose per fraction d_L or:

$$\mathrm{RBE} = \frac{-(\alpha/\beta)_L + \sqrt{(\alpha/\beta)_L^2 + 4d_H\left[(\alpha/\beta)_L \mathrm{RBE}_{max} + \mathrm{RBE}_{min}^2 d_H\right]}}{2d_H} \tag{2.7}$$

when expressed in terms of the high LET dose per fraction d_H.

Figure 2.4 was derived using $\mathrm{RBE}_{max} = 5$, $\mathrm{RBE}_{min} = 1$ and $(\alpha/\beta)_L = 3$ Gy, but the general trend of a steadily falling RBE with increasing dose per fraction is independent of the chosen values. Clearly, the assumption of a fixed value of RBE, if applied to all fraction sizes, could lead to gross clinical errors and Eqs (2.6) and (2.7) make the point that determination of RBEs in a clinical setting is potentially complex and will depend on accurate knowledge of RBE_{max} and (if it is not unity) RBE_{min}. Although there is not yet clear evidence over whether or not there is a consistent trend for RBE_{min} to be non-unity, the possibility is nevertheless important as it may hold very significant implications.

Figure 2.4 also shows schematically how the rate of change of RBE with changing dose per fraction is influenced by the existence of a non-unity RBE_{min} parameter. Even for a fixed value of RBE_{max}, the potential uncertainty in the RBE values at the fraction sizes likely to be used clinically might themselves be very large if RBE_{min} is erroneously assumed to be unity. These uncertainties would be compounded if there were an additional linkage between RBE_{max} and the tissue α/β value.

As is seen from Eqs (2.6) and (2.7), the RBE value at any particular dose fraction size will also be governed by the low LET α/β ratio (a tissue dependent parameter which provides a measure of how tissues respond to changes in dose fractionation) and the dose fraction size (a purely physical parameter) at the point under consideration. Finally, and as has been shown through the earlier clinical experience with neutron therapy, the RBE_{max} value may itself be tissue dependent, likely being higher for the dose-limiting normal tissues than for tumours. This tendency is borne out by experimental evidence using a variety of ion species as well as by theoretical microdosimetric studies. This potentially deleterious effect may be offset by the fact that, in carbon-, helium- and argon-ion beams, LET (and, hence, RBE) will vary along the track in such a way that it is low at the entry point (adjacent to normal tissues) and highest at the Bragg peak located in the tumour. However, although this might be beneficial, it does mean that the local RBE is more spatially variable than is indicated by Eq. (2.6).

Owing to the difficulties in setting reference doses at which clinical inter-comparisons could be made more straightforward, Wambersie proposed that a distinction be made between the ‘reference’ RBE and the ‘clinical’

RBE. Thus, the reference RBE might be that determined at 2 Gy fractions on a biological system end point representative, for example, of the overall late tolerance of normal tissues. As more clinical experience of using the particular radiation becomes available, a more practical 'clinical' RBE evolves, this being the reference RBE empirically weighted by collective clinical experience and by volume effects related to the beam characteristics, geometry or technical conditions.

2.6.5. The dose rate effect and the concept of repeat treatments

When mammalian cells are irradiated, it is helpful to visualize their subsequent death as resulting from either of two possible processes. In the first process, the critical nuclear target (DNA) is subjected to a large deposition of energy which physically breaks both strands of the double helix structure and disrupts the code sufficiently to disallow any opportunity of repair. This process can be thought of as a single-hit process and the total amount of DNA damage created this way is directly proportional to the dose delivered.

In the second process, an ionizing event occurs and releases only sufficient energy to disrupt the coding carried by one strand of the DNA. Following this event, and if the irradiation continues, two outcomes are possible: either the broken strand will restore itself to its original state (no lethality) or, prior to full repair taking place, a second, independent radiation event may occur in the same location and damage the opposite strand of the DNA, a complementary action between the two damaged strands then leading to cell lethality in what is called a two-hit process. Since this route depends on there being two independent events, each having a probability proportional to dose, the number of damaged DNA targets created this way is proportional to dose × dose, i.e. dose^2. Once created, the radiation damage due to these two possible routes is indistinguishable (i.e. both processes are lethal). From this simplified description, it is clear that the observed radiation response characterized in the cell survival curve will consist of two components: one linear with dose and the other quadratic, i.e. proportional to dose^2. This phenomenological description qualitatively explains the shape of a radiation survival curve, with a finite initial slope at low dose followed by an increasingly downward curvature as dose increases.

However, the amount of damage created in the second process is dependent on the ability of the second break to be induced before the first break has repaired itself and, thus, is dependent on the dose rate.

Figure 2.5 shows a range of response curves in which the doses are delivered at four different dose rates, the individual doses taking proportionately longer to deliver as dose rate is reduced. This graph illustrates that reducing the dose rate causes the overall shape of the response curve to become less 'curvy'

than in the acute case, but that the initial slope remains unchanged. When the doses are all delivered at a very low dose rate, as is the case for most radionuclide therapies, the response is essentially a straight line, when the curves are plotted on a log-linear scale, as is common practice for radiation survival curves. This means that the low dose response is purely exponential.

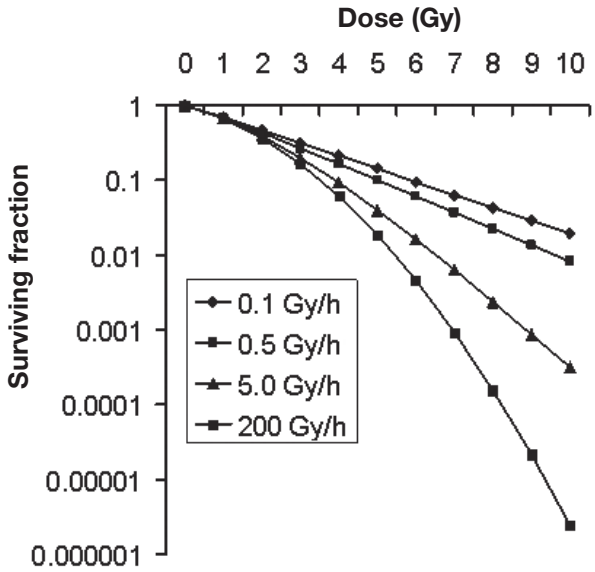

FIG. 2.5. Surviving fraction as a function of dose for different dose rates. It is important to note that most radionuclide therapies are delivered at low dose rate in the range of 0.1–0.5 Gy/h, when the survival curve is almost linear.

2.6.6. The basic linear–quadratic model

The basic equation describing the shape of the cell survival curves shown in Fig. 2.2 is referred to as the LQ model, which has a biophysical origin. Cell survival following delivery of an acute dose d is given as:

$$S = \exp\left(-\alpha d - \beta d^2\right) \tag{2.8}$$

where α (in units of Gy^{-1}) and β (in units of Gy^{-2}) are the respective linear and quadratic sensitivity coefficients.

If the treatment is repeated in N well spaced fractions, then the net survival is S_N, where:

$$S_N = S^N = \exp\left(-N\alpha d - N\beta d^2\right) \tag{2.9}$$

Taking natural logarithms on both sides of Eq. (2.9) and dividing throughout by α leads to:

$$\frac{\ln S_N}{\alpha} = -Nd - \frac{Nd^2}{(\alpha/\beta)} \tag{2.10}$$

2.6.7. Modification to the linear–quadratic model for radionuclide therapies

Targeted radionuclide therapy normally involves irradiation of the tumour/normal tissues at a dose rate which is not constant but which reduces as treatment proceeds, as a consequence of the combination of radionuclide decay and biological clearance of the radiopharmaceutical. To allow for this, a more extensive formulation of the LQ model is required.

2.6.8. Quantitative intercomparison of different treatment types

In many aspects of LQ modelling, a term called the 'biological effective dose' (BED) is employed to assess and inter-compare different treatment types. BED is defined as:

$$\text{BED} = -\frac{\ln S_N}{\alpha} = Nd\left[1 + \frac{d}{(\alpha/\beta)}\right] \tag{2.11}$$

Although the parameters α and β are rarely known in detail for individual tumours or tissues, values of the ratio α/β (in units of grays) are becoming increasingly known from clinical and experimental data. In general, α/β is systematically higher (5–20 Gy) for tumours than for critical, late-responding normal tissues (2–5 Gy) and it is this difference which provides the BED concept with much of its practical usefulness.

For non-acute treatments (those in which the dose delivery is protracted over a long time period on account of a lower dose rate), the BED is re-written as:

$$\mathrm{BED} = Nd\left[1+\frac{d\ g(t)}{(\alpha/\beta)}\right] \tag{2.12}$$

where $g(t)$ is a function of the time t taken for delivery:

$$g(t) = \frac{2}{\mu}\left[1-\frac{1-\exp(-\mu t)}{\mu t}\right] \tag{2.13}$$

and where μ is the mono-exponential time constant relating to the repair of sublethal damage. μ is related to the tissue repair half-time ($T_{1/2}$) via:

$$\mu = \frac{0.693}{T_{1/2}} \tag{2.14}$$

For a treatment delivery at constant dose rate R, the delivered dose d is related to treatment time t via $d = Rt$, thus:

$$\mathrm{BED} = Rt\left[1+\frac{2R}{\mu(\alpha/\beta)}\left\{1-\frac{1-\exp(-\mu t)}{\mu t}\right\}\right] \tag{2.15}$$

When $t > 12$ h, Eq. (12.15) simplifies to:

$$\mathrm{BED} = Rt\left[1+\frac{2R}{\mu(\alpha/\beta)}\right] \tag{2.16}$$

2.6.9. Cellular recovery processes

At lower doses and dose rates, cellular recovery may play an important role in the fixation of the radiation damage. There are three broad types of cellular radiation damage:

(a) Lethal damage in which the cellular DNA is irreversibly damaged to such an extent that the cell dies or loses its proliferative capacity;
(b) Sublethal damage in which partially damaged DNA is left with sufficient capacity to restore itself over a period of a few hours, provided there is no further damage during the repair period;

(c) Potentially lethal damage in which repair of what would normally be a lethal event is made possible by manipulation of the post-irradiation cellular environment.

2.6.10. Consequence of radionuclide heterogeneity

The effectiveness per unit dose of a radiopharmaceutical depends on the heterogeneity of the radionuclide distribution. Global non-uniformity of a source distribution, which results in pockets of cells (tumour or normal tissue) receiving less than the average dose will almost always result in a greater fraction of cell survivors, than if all cells receive a uniform dose. The one possible exception would be if a radiopharmaceutical would selectively localize at sensitive target cells, within an organ, that are key for organ regeneration or function, e.g. crypt cells in the colon. The cellular response also depends on the microdosimetry, especially if the radiopharmaceutical in question selectively localizes on the cell surface or internalizes within a certain cohort of cells within a tumour/normal organ. Radiolabels that selectively localize on the surface of cells or are internalized may exhibit geometric enhancement factors that modulate a response. The reader is referred to ICRU Report 67 on absorbed dose specification in nuclear medicine for more details.

2.7. GROSS RADIATION EFFECTS ON TUMOURS AND TISSUES/ORGANS

2.7.1. Classification of radiation damage (early versus late)

Cells which are lethally affected by radiation may continue to function for some time after the infliction of the damage, only dying when attempting to undergo subsequent cell division (mitosis). Clinically observed radiation effects in whole tissues or organs reflect the damage inflicted to large numbers of constituent cells and, thus, appear on a timescale which is governed largely by the underlying proliferation rates of those cells. Such observable effects are classified as being either late or early, depending on the speed at which they manifest themselves following irradiation. Late effects appear months or years after irradiation and appear in structures which proliferate very slowly, e.g. kidney. Early (or acute) effects appear within days, weeks or months of irradiation and are associated with fast-proliferating epithelial tissues, e.g. bone marrow, mucosa, intestinal tract, etc.

In most types of radiotherapy, it is the late effects which are considered to be most critical and which generally limit the total dose which may be delivered to the tumour. If the radiation tolerance of the late-responding tissues is exceeded, then the subsequent reactions, depending on the tissues in which they arise, may seriously affect mobility and/or quality of life, and may even be life threatening. Such problems arise long after the completion of treatment and are, thus, impossible to correct. These are the serious considerations which are at the heart of the therapeutic index concept discussed below (see Section 2.7.3). Acute reactions in radiotherapy, although they may be unpleasant, are usually transient and easier to control by adjustment of the treatment dose delivery pattern and/or simple medication. In radionuclide therapies, it is in most instances possible to circumvent acute radiation toxicities once they begin to occur, such as by accelerating clearance of the radiopharmaceutical. Chronic toxicities, such as to the kidney, usually occur at times which are long relative to the lifetime of the radionuclide. Hence, considerable importance should be attributed to the administration of safe activities of therapeutic radionuclides that do not exceed any dose limiting constraints.

2.7.2. Determinants of tumour response

Irrespective of the mechanism used to achieve tumour targeting, the potential advantage of radionuclide therapy over other forms of radiation therapy is its ability to deliver dose to both the local disease and to occult tumour deposits.

In nuclear medicine, the primary determinants of treatment effectiveness are:

— The tumour specificity of the radionuclide carrier.
— The homogeneity of uptake of the carrier within the targeted tumour(s).
— The intrinsic RBE (see Section 2.6.4) of the radiation used for the therapy: this is determined primarily by the nature of the radionuclide emissions (e.g. α particles, β particles, low energy γ rays, Auger electrons, etc.).
— The range of the particles, as determined by their energies.
— The total dose delivered.
— The responsiveness of the targeted tumour cells to radiation. This will be determined by radiobiological properties such as cellular radiosensitivity and the variations of sensitivity within the cell cycle, the oxygen status of the cells (fully oxic, partially oxic or hypoxic), the ability of the cells to recover from sublethal radiation damage and the degree to which tumour growth (repopulation) may occur during therapy.

These factors are complementary and interactive, and should not be considered in isolation from each other. Thus, for example, significant non-uniformity of uptake within the tumour may result in dose 'cold spots', but the detrimental potential of these might be offset by the selection of a radionuclide which emits particles of sufficient range to produce a cross-fire effect within the cold spots from those adjacent cells which are properly targeted. The significance of cold spot and cross-fire effects is further dependent on the size of the tumour deposit under consideration.

2.7.3. The concept of therapeutic index in radiation therapy and radionuclide therapy

The therapeutic index of a particular radiation treatment (often referred to in older publications as the 'therapeutic ratio') is a measure of the resultant damage to the tumour vis a vis the damage to critical normal structures. Treatments with a high therapeutic index will demonstrate good tumour control and low normal tissue morbidity; treatments with a low therapeutic index will be associated with a low tumour control and/or high morbidity. There have been several attempts to provide quantitative definitions of therapeutic index, but it is usually sufficient to consider therapeutic index as being a qualitative concept — any new treatment which, relative to an existing treatment, improves tumour control and/or reduces morbidity is said to be associated with an improved therapeutic index.

In conventional (external beam) radiotherapy, the normal tissues at risk will be those immediately adjacent to the tumour being treated. Doses to the normal tissues (along with the risk of toxicity) may be reduced by attention to a combination of physical and radiobiological factors. In targeted radionuclide therapy, the tumour may be single and discrete (as is the case in most external beam therapy) or may consist of distributed masses or metastatic deposits at several locations within the body. The normal tissues at risk may themselves be widely distributed but, more particularly, may be a reflection of the particular uptake pattern of the targeting compound being used for the therapy.

2.7.4. Long term concerns: stochastic and deterministic effects

The radiation detriment which results from radiation exposure may be classified as being either stochastic or deterministic in nature. Stochastic effects (e.g. hereditary damage, cancer induction) are those for which the likelihood of them occurring is dose related, but the severity of the resultant condition is not related to the dose received. Deterministic effects (e.g. cataract induction, general radiation syndromes, bone marrow ablation, etc.) manifest themselves with a severity which is dose related. In general, it is predominantly stochastic

effects which need to be considered as potential side effects from diagnostic uses of radionuclides, although deterministic damage may result if the embryo or fetus is irradiated. For radionuclide therapy applications, the concerns relate to both stochastic and deterministic effects.

2.8. SPECIAL RADIOBIOLOGICAL CONSIDERATIONS IN TARGETED RADIONUCLIDE THERAPY

2.8.1. Radionuclide targeting

Tumour targeted radiotherapy is a very promising approach for the treatment of wide-spread metastasis and disseminated tumour cells. This technique aims to deliver therapeutic irradiation doses to the tumour while sparing normal tissues by targeting a structure that is abundant in tumour cells, but rare in normal tissues. This can be done by using antibodies labelled with a therapeutic relevant radionuclide acting against a specific tumour target. Radiolabelled antibody therapy has already become common in the treatment of non-Hodgkin's lymphoma, e.g. ^{131}I-tositumomab (Bexxar®) and ^{90}Y-ibritumomab tiuxetan (Zevalin®), and exhibits great potential for being extended to other diseases. A good example is epidermal growth factor (EGF) labelled with ^{125}I which will bind EGF receptors. EGF receptors are overexpressed on tumour cells in many malignancies such as highly malignant gliomas. At present, several other radiolabelled antibodies are being used in experimental models and in clinical trials to study their feasibility in other types of cancer.

2.8.2. Whole body irradiation

Conventional external beam radiotherapy involves controlled irradiation of a carefully delineated target volume. Normal structures adjacent to the tumour will likely receive a dose, in some cases a moderately high dose, but the volumes involved are relatively small. The rest of the body receives only a minimal dose, mostly arising from radiation scattered within the patient from the target volume and from a small amount of leakage radiation emanating from the treatment machine outside the body.

Targeted radionuclide therapies are most commonly administered intravenously and, thus, can give rise to substantial whole body doses and, in particular, doses to the radiation sensitive bone marrow. Once the untargeted activity is removed from the blood, it may give rise to substantial doses in normal structures, especially the kidneys. Furthermore, the activity taken up by

the kidneys and targeted tumour deposits may (if γ ray emissions are involved) continue to irradiate the rest of the body.

2.8.3. Critical normal tissues for radiation and radionuclide therapies

Since the radiation doses used in radionuclide therapies are much higher than the doses used for diagnosis, (prolonged) retention of the pharmaceuticals within the blood circulation and, hence, increased accumulation of radionuclides in non-tumour cells, might lead to unwanted toxicities. The bone marrow, kidney and liver are regarded as the main critical organs for systemic radionuclide therapy. Other organs at risk are the intestinal tract and the lungs. The bone marrow is very sensitive towards ionizing radiation. Exposure of the bone marrow with high doses of radiation will lead to a rapid depression of white blood cells followed a few weeks later by platelet depression, and in a later stage (approximately one month after exposure) also by depression of the red blood cells. In general, these patients could suffer from infections, bleeding and anaemia. Radiation damage to the gastrointestinal tract is characterized by de-population of the intestinal mucosa (usually between 3 and 10 days) leading to prolonged diarrhoea, dehydration, loss of weight, etc. The kidneys, liver and lungs will show radiation induced damage several months after exposure. In kidneys, a reduction of proximal tubule cells is observed. These pathological changes finally lead to nephropathy. In the liver, hepatocytes are the radiosensitive targets. Since the lifespan of the cells is about a year, deterioration of liver function will become apparent between 3 and 9 months after exposure. In lungs, pulmonary damage is observed in two waves: an acute wave of pneumonitis and later fibrosis.

The determinants of normal tissue response from radionuclide studies is a large subject due to the diversity of radiopharmaceuticals with differing pharmacokinetics and biodistribution, and the widely differing responses and tolerances of the critical normal tissues. A principal determinant of the type of toxicity depends on the radionuclide employed. For example, isotopes of iodine localize in the thyroid (unless blocked), salivary glands, stomach and bladder. Strontium, yttrium, samarium, fluorine, radium, etc. concentrate in bone. Several radiometals, such as bismuth, can accumulate in the kidney. If these radionuclides are tightly conjugated to a targeting molecule, the biodistribution and clearance are determined by that molecule. For high molecular weight targeting agents, such as an antibody injected intravenously, the slow plasma clearance results in marrow toxicity being the principal dose limiting organ. For smaller radiolabelled peptides, renal toxicity becomes of concern. When studying a new radiopharmaceutical or molecular imaging agent, it is always important to perform a detailed study of the biodistribution at trace doses, to ensure the

absence of radionuclide sequestration within potentially sensitive tissue, such as the retina of the eye or the germ cells of the testes.

A review of normal tissue toxicities resulting from radionuclide therapies is given by Meredith et al. (2008).

2.8.4. Imaging the radiobiology of tumours

The development of molecular imaging using positron emission tomography (PET) has given rise to new radiotracers which have the potential to assess several features of radiobiological relevance for therapy planning. One tracer that is becoming more widely available for PET imaging is fluorothymidine. This radiotracer exhibits the property of becoming selectively entrapped within cells that are progressing through S-phase (DNA replication) of the cell cycle, thus providing a signal which should be proportional to cell proliferation, and minimizing the signal from cells in G_0 or in cell cycle arrest. The ability to selectively identify only replicating cells separate from all tumour cells present within the computed tomography-determined tumour volume may present an excellent opportunity for more accurate measures of the initial viable tumour burden as well as evaluating tumour response. Complementary to measuring tumour response is the measurement of therapeutic efficacy through radiotracers that selectively target cell death. Radiotracers are under development with the ability to selectively bind to receptors expressed on cells undergoing programmed cell death, e.g. radiolabelled annexin V. Another area of active research is in the field of hypoxia imaging. Cells within a tumour microenvironmental region of low partial oxygen pressure, i.e. hypoxia, are known to exhibit a great radio-resistance to both radiation and chemotherapy relative to those under normoxic conditions. A number of PET radiotracers are under evaluation for imaging tumour hypoxia with PET, including fluoromisonidazole (^{18}F-FMISO), fluoroazomycin arabinoside (^{18}F-FAZA) and copper-diacetyl-bis(N4-methylthiosemicarbazone) (^{64}Cu-ATSM). The ability to measure the radiobiological attributes of a tumour prior to therapy may provide invaluable information concerning the relative resistance/aggressiveness of tumours, leading to improved management of these patients.

2.8.5. Choice of radionuclide to maximize therapeutic index

The choice of the optimum radionuclide to maximize the therapeutic index depends on a number of factors. First, the range of the emitted particles from the radionuclide should depend on the type of tumour being treated. For leukaemia or micrometastatic deposits, consisting of individual or small clusters of tumour cells, there is a distinct advantage of using radionuclides which emit very short

range particles. Since α particles have ranges of <100 μm in tissue, α particle emitters would have an advantage, if the targeting molecule were able to reach all tumour cells. However, α particle emitting radionuclides are not widely available and are extremely expensive. In addition, the short range of α particles can be a disadvantage for bulk tumours. For these reasons, almost all therapeutic radionuclides utilized in the clinic today consist of medium (^{131}I) or long range (^{90}Y, ^{186}Re) β emitters. These radionuclides are advantageous when treating solid tumours for which target receptor (antigen) expression may be heterogeneous, or with non-uniform delivery, due to the greater cross-fire range of their β emissions (ranging up to a 1 cm range in unit density tissue).

A second important consideration is the choice of radionuclide half-life. If the half-life is too short, then the radiolabelled tumour targeting agent may have insufficient time to reach its target, resulting in a minimal therapeutic index. Increasing the half-life will increase the therapeutic index, but render the patient radioactive for a longer period of time, resulting in prolonged confinement, greater expense and radiation risks to staff and family. Pure β emitting radionuclides such as ^{90}Y and ^{32}P have advantages in that they minimize the exposure to personnel assisting the patient. The half-life of the radionuclide should ideally match the biological uptake and retention kinetics of the tumour-targeting carrier used. For large protein carriers such as antibodies, radionuclides with half-lives of several days are required to optimize the therapeutic index. For smaller molecular targeting agents such as peptides, short lived radionuclides may be better suited to minimize radioactive waste.

Thirdly, it is necessary to consider radiochemistry, ease and stability of the radiolabelled end product. All of these factors need to be taken into consideration in order to produce the optimum therapeutic targeting compound for use in clinical therapeutic applications.

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