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Uncertainties in the relative biological effectiveness of therapeutic proton beams associated with bias towards high doses per fraction in radiobiological experiments

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Scientific Note

Abstract

Most data supporting the widely accepted relative biological effectiveness (RBE) value of 1.1 for therapeutic proton beams are from radiobiological experiments with relatively high doses per fraction. The purpose of this study was to estimate bias in RBE that differences in dose levels between these experiments and proton radiotherapy treatments may cause. The linear quadratic model was applied to calculate, using prior experimental data, RBE variations with dose and α/β ratio for doses delivered in a standard fractionation regimen. The results suggest that the RBE measured at relatively high doses per fraction typical for a radiobiological experiment underestimates the RBE of proton radiotherapy with a standard fractionation. The bias increases with decreasing radiation dose and decreasing α/β ratio, suggesting that, if differences in dose levels are not accounted for, there may be a large underestimation of biological effects in late-responding tissues exposed to low doses of radiation.

Keywords: Proton beam therapy, Proton RBE, Hadron therapy, Dose fractionation

1. Introduction

In proton radiotherapy, a generic proton relative biological effectiveness (RBE) value of 1.1 is widely accepted.[1] However, several studies investigating the biological effects of proton and photon beams at the molecular and cellular levels have reported large differences between the two types of radiation.[2-4] These results reflect important differences in the patterns of molecular damage induced by protons and photons and in the mechanisms by which that damage is repaired. They also suggest that, for some biological endpoints, the proton RBE value can be significantly greater than one.

An RBE value of 1.1 is consistent with the average RBE value calculated in a comprehensive review of pertinent in vivo studies.[5] Those calculations, however, were affected by the limitations of the radiobiological data available at that time. For example, the experimental RBE data used in those calculations were dominated by only four biological endpoints, all from murine studies: survival after thoracic irradiation, inactivation of intestinal crypt cells, acute skin reactions, and inactivation of fibrosarcoma cells. In addition, the data were heavily biased toward early-responding, high-α/β-ratio tissues. Furthermore, most of the RBE measurements were made at locations within the spread-out Bragg peak. Therefore, the linear energy transfer (LET) spectra were similar to the LET spectra in the target volume covering the tumor but different from the LET spectra in the normal tissues outside the target volume. Finally, the doses per fraction in those experiments were typically much higher than the standard fraction dose of approximately 2 Gy, exceeding 10 Gy per fraction in most cases. Measurements taken directly at doses of 2 Gy or less constitute only about one-tenth of the sample. Consequently, the use of the average RBE derived from these data is associated with less uncertainty in hypofractionated treatments than in standard fractionation treatments.

In summary, a proton RBE of 1.1 is supported by limited and skewed radiobiological data and is most appropriate for assessment of tumor control (but not normal tissue complication probability) in hypofractionated treatments, excluding malignancies characterized by relatively low α/β ratios, such as prostate cancer.[6] To derive more confident estimates of proton RBE outside the stated scope, additional radiobiological data are

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needed. These data should be collected under conditions approximating those of specific types of treatment in terms of dose ranges, fractionation schemes, and tissue types.

Published in vitro data provide some indication of the magnitude of possible bias in the estimated proton RBE value. Dose per fraction in RBE measurements in vitro tends to be lower than in measurements in vivo, as can be seen from the data compiled by Paganetti *et al*.[5] Also, this compilation included in vitro data with relatively low α/β ratios. These observations may help explain the difference in the reported average in vivo and in vitro RBE values in Paganetti *et al*.'s study (1.10 and 1.22, respectively). In addition, Gerweck and Kozin[7] demonstrated a particularly strong dose dependence of RBE for V79 cells. In V79 cell line, the proton RBE value was 30% higher at the cell survival level that corresponded to a photon dose of approximately 2 Gy than at the level that corresponded to a photon dose of 6 Gy. At approximately 2 Gy, the proton RBE level was rather high (about 1.6).

Paganetti *et al*. noted an increased RBE value at lower doses per fraction.[5] An earlier in vivo study reported more specifically that "RBEs were found to increase slightly (about 4%) with decreasing dose, in the investigated dose range (12-17 Gy)".[8] However, in Paganetti *et al*.'s study the increase in RBEs was apparently deemed negligible, at least for in vivo systems.[5] The lack of a clear trend in that data set likely reflects the fact that the data are mostly from experiments with early responding tissues, as mentioned earlier. Therefore, the trend toward higher RBE values, if any, would be expected to be weak. With typical experimental uncertainties of 5-10%, and which are much higher in some cases, any trend would be difficult to identify. On the other hand, the fractionation schedule in radiotherapy is known to have a profound effect on treatment outcomes. Furthermore, even relatively small RBE variations, which are difficult to detect and quantify in in vivo experiments, may have a statistically significant impact on a population of radiotherapy patients. Given the limitations of experimental data, there is a clear need for a theoretical analysis. The linear quadratic model is an obvious choice for such an undertaking, since it has been used in clinical studies for decades, in particular for the analysis and design of fractionation schedules.[9]

In the present study, the linear quadratic model was applied to previously reported data on in vivo RBE[5] to estimate RBE variations due to variations of radiation dose and tissue α/β ratios. To my knowledge, no such analysis has been previously reported, although it has been previously indicated that the RBE may increase with decreasing dose and/or decreasing α/β ratios.[7] The present study addresses this possible problem quantitatively using a general formalism. The purpose of the study was to estimate the bias and uncertainties in proton RBE for the standard radiotherapy fractionation schedule when it is derived from radiobiological experiments performed at doses per fraction higher than those in the radiotherapy treatments, without correction for differences in fraction size. The study was also intended to assess the magnitude of potential errors, identify major trends, and provide preliminary data that may aid in the design of future experiments. The approach, however, is prone to large uncertainties due to the limitations of the available experimental data. Therefore, the results reported in this study cannot be applied directly to alter current proton radiotherapy practices.

2. Methods and Materials

This section describes a simple method of calculating the RBE for clinically relevant dose fractionation schemes using RBE data generated from arbitrary doses or doses-per-fraction experiments. The method is based on the linear quadratic model. Let d_γ and d_p be doses per fraction delivered to a point of interest within an organ or tissue, where the subscripts γ and p refer to photon and proton radiation fields, respectively. To adhere to the conventions of proton therapy, the reference photon field here is that produced by a ^{60}Co source. This source is widely used in radiobiological experiments. The numbers of photon and proton dose fractions are n_γ and n_p, respectively. The parameters of the model are α_γ, β_γ, α_p, and β_p. The proton RBE is the ratio of the total photon and proton doses, $D_\gamma = n_\gamma d_\gamma$ and $D_p = n_p d_p$, that cause equal biological effect. In the standard basic form of the linear quadratic model, the equal effect condition is given by this equation[9]:

$$n_\gamma\left(\alpha_\gamma d_\gamma + \beta_\gamma d_\gamma^2\right) = n_p\left(\alpha_p d_p + \beta_p d_p^2\right). \qquad (1)$$

This equation does not include any time-dependent factors to account for effects related to cell kinetics, such as cell proliferation. Calculating the impact of cell kinetics on the RBE is outside the scope of this study.

To reduce the number of parameters, two additional assumptions are made. The first assumption is that the number of fractions is the same for both photon and proton irradiations, *i.e*., $n_\gamma = n_p$. This condition is typically met in radiobiological experiments designed to measure the proton RBE. The second assumption is that parameter β does not depend on radiation quality, *i.e*., $\beta_\gamma = \beta_p$. This assumption is supported by the theory of dual radiation action that attributes the quadratic term to the intertrack interactions of radiation-induced sublesions.[10] A study by Coutrakon *et al*.[11] provides a good example of experimental data on the dependence of parameters α and β on radiation quality. Coutrakon *et al*. measured cell survival curves at multiple depths within typical therapeutic proton beams at three

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different beam energies. The ratio β_p/β_γ ranged from 1.0 to 1.4 with a median of 1.2. In contrast, the ratio α_p/α_γ was much larger, with values ranging from 2.0 to 3.3 and a median of 2.5. For high LET radiation of approximately 30-100 keV/μm, experimental data show a dependence of parameter β on LET.[12] For protons of therapeutic energies, experimental data on variations of β with radiation quality, or the lack thereof, are far less conclusive, especially in vivo. It is therefore a sensible approach to follow theoretical arguments suggesting that $\beta_\gamma = \beta_p$.[10] This assumption is also consistent with the trend of the RBE approaching a value of one at high doses, a tendency which, for example, is observed in fast neutrons.[13] A more elaborate model for β would be difficult to justify.

The final formula used in my calculations is given below. It was derived from Eq. (1) under the stated assumptions:

$$R_2 = \frac{1}{2b}\left\{\sqrt{1+4bR_1+4b^2\left[1+\left(R_1^2-1\right)\frac{d_{p1}}{d_{p2}}\right]}-1\right\}, \qquad (2)$$

Here R_1 and R_2 are RBEs measured at doses d_{p1} and d_{p2}, and $b = \beta\, d_{p2} / \alpha_\gamma$ is a dimensionless variable.

Qualitatively, in this formalism the RBE increases with decreasing dose because α is more affected by radiation quality than is β. At sufficiently high doses, the RBE tends to 1, which, as we have mentioned, is consistent with fast neutron data for several endpoints[13] and is a mathematical corollary of the assumption $\beta_\gamma = \beta_p$.

Table 1: RBE dependence on photon dose per fraction: comparison of other researchers' measurements[14-16] with present calculations.

Reference	Dose, Gy	RBE measure-ments	$\alpha_\gamma / \beta_\gamma^*$ Gy	Dose, Gy	RBE		Dose, Gy	RBE	
					Prior measurements	Present calculations		Prior measurements	Present calculations
Blomquist *et al.*[14]	9.9	1.15	3.65	6.5	1.28 ± 0.21	1.22	3.0	1.63 ± 0.63	1.41
Tang *et al.*[15]	8.2	1.12	6.52	3.9	1.18 ± 0.04	1.19	1.2	1.27 ± 0.05	1.30
Wouters *et al.*[16]	10.0	1.19	5.00	3.8	1.28	1.37	1.9	1.49	1.54

* $\alpha_\gamma / \beta_\gamma$ used in the calculations

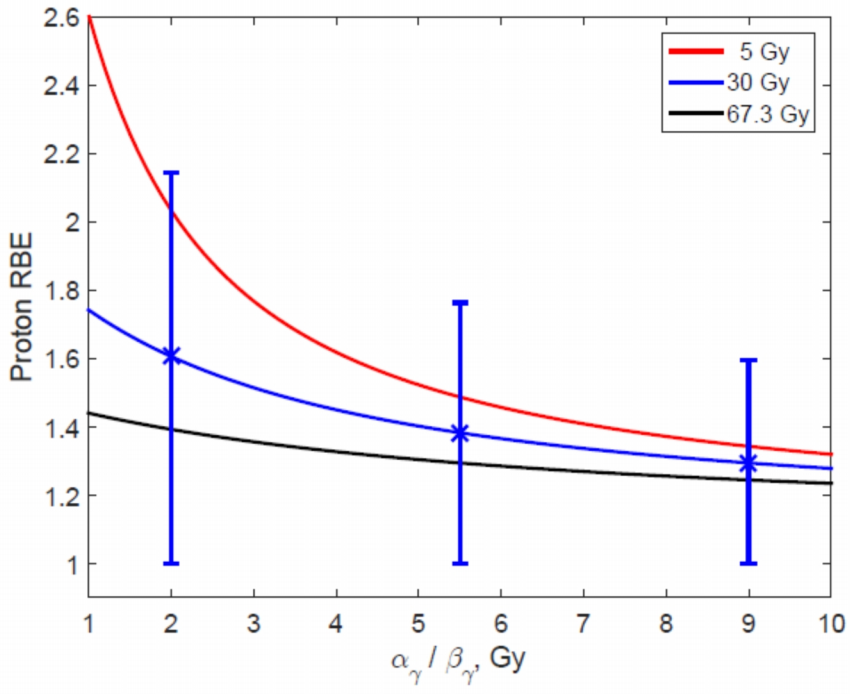

Figure 1: Calculated proton RBE versus $\alpha_\gamma / \beta_\gamma$ ratio for total proton doses of 5, 30, and 67.3 Gy delivered in 37 fractions. Black line, 67.3 Gy; blue line, 30 Gy; and red line, 5 Gy. The confidence intervals are shown for the dose of 30 Gy and $\alpha_\gamma / \beta_\gamma$ = 2, 5.5, and 9 Gy.

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3. Results and Discussion

To test the described RBE calculation method, I used data from in vitro studies that reported proton RBEs for cell survival at several survival levels.[14-16] The lowest survival levels tested in those previous studies were at photon doses per fraction of approximately 8-10 Gy. We used the $\alpha_\gamma/\beta_\gamma$ ratios and RBEs at the lowest survival levels reported in those studies to calculate the RBE at lower doses. The analysis was limited to measurements made approximately in the middle of a spread-out Bragg peak. Wouters *et al*. fitted the cell survival data for photons into a two-population, linear quadratic model.[16] The measured RBEs presented in Table 1 for that study were derived from the two-population model because it most closely fits the measured points. The $\alpha_\gamma/\beta_\gamma$ ratio needed to apply my formalism was taken from a separate fit in the high-dose region because the purpose of these calculations was to extend high-dose RBE data to the low-dose range. My calculated RBEs at low doses are compared in Table 1 with the prior measurements at the same doses.[14-16] Agreement between our calculations and the published measurements of Blomquist *et al.* and Tang *et al*.[14,15] was within experimental uncertainties. Discrepancies between the present calculations and the data reported by Wouters *et al*.[16] may have exceeded experimental uncertainties (these were reported in a graph only). The differences, however, were within 10%. The discrepancies were caused by deviations from our assumption that $\beta_\gamma = \beta_p$ and, in the case of the photon measurements reported by Wouters *et al*.[16], by deviations from the simple linear quadratic model.

The agreement of my model with in vivo data was investigated using a subset of the data compiled by Paganetti *et al*[5], where RBEs were reported at doses of 2 Gy or less. This subset is comprised of four studies: three from Harvard University[17-19] and one, more recent, from the National Accelerator Center in South Africa.[20] Two of the studies[17,20] reported on the survival of jejunum intestinal crypt cells in mice. In both of these studies, the dose-effect dependences were fitted with the linear model, *i.e.* $\beta_\gamma = \beta_p = 0$. The reported RBE values ranged from 1.14 to 1.23, and the number of fractions ranged from 1 to 20. The RBE value in these studies did not vary significantly with fractionation. This finding is consistent with our model applied to the special case of $\beta_\gamma = \beta_p = 0$. Furthermore, Gueulette *et al* concluded that the "clinical RBE value of 1.10 appears low."[20]

An RBE value of 1.16 ± 0.12 (average ± standard deviation) was reported at a cell survival level of 0.5 for a spontaneous murine tumor.[18] The $\alpha_\gamma/\beta_\gamma$ ratios were in the range of 8.0 - 43 Gy, with a median of 24 Gy. I used α_γ, β_γ, and RBEs at the survival level of 0.1 reported in Urano *et al*.'s study and applied my model to calculate RBE values at a survival level of 0.5. The calculated average RBE, 1.22 ± 0.10, was higher than that reported by Urano *et al*[18], but not significantly. The maximum number of fractions studied (Urano *et al* [18]) was 10. In that case the measured[18] and calculated RBE values were 1.42 and 1.36, respectively. It should be noted that the data from these three studies concern only early-responding tissues and that the parameters of the linear quadratic model had large uncertainties. For example, in two of Urano *et al*.'s duplicate, single-fraction experiments, the $\alpha_\gamma/\beta_\gamma$ ratios differed by a factor of 2.5.

Finally, the second study by Urano *et al*[19] reported RBE values for weight loss in mouse testes, measured 35 days after irradiation. The animals were irradiated with 0.5 to 5 Gy, administered to their whole body. At the level of 20% weight loss relative to non-irradiated control animals, the RBE value was 1.21 ± 0.08. The photon dose at this effect level was 0.7 Gy. The weight loss dose dependence was best described by a linear function of the dose logarithm, and therefore the data are outside the scope of the linear quadratic model. Overall, except this last example, our model was compatible with prior experimental data. These data, however, are not sufficient to allow for confident confirmation or rejection of the model. Experimental data for a broader range of $\alpha_\gamma/\beta_\gamma$ ratios are needed for that. It must be added that Paganetti *et al*.'s study[5] disputed the dosimetry of these earlier experiments.[17-19] Accordingly, the RBE values reported in these earlier studies were adjusted accordingly resulting in lower values. This correction was justified by a report addressing a problem with a clinical dosimetry protocol at the Harvard Cyclotron Laboratory that was based on Faraday Cup methods for absolute dose calibration.[21] On the other hand, the methodology described in the original publications[17-19] appears to be adequate because a calorimeter used for "an independent check of the dosimetry agreed with the standard dosimetry … within 0.8 to 2.6%".[22] For this reason, I have quoted unadjusted RBE values in this paper.

I also applied my model to an analysis of the in vivo data summarized by Paganetti *et al*.[5] The median photon dose per fraction for experiments in which the RBE value was measured in the middle of a spread-out Bragg peak was 12 Gy. The median RBE value was 1.1, and the standard deviation of the RBE was 0.1. On the basis of these observations, I assumed that the RBE of 1.1 was measured at 12 Gy per fraction. This assumption identified a single data point representative of a typical for this data set experiment that I chose to demonstrate the potential effects of fractionation on the RBE. I then used this data point and my formalism to calculate the RBE value for a standard fractionation schedule of 37 fractions with total doses of 5, 30, and 67.3 Gy. In this calculation, the number of fractions and the highest dose represent a typical prescription for proton radiotherapy

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for prostate cancer[23], and the lower doses are relevant for normal tissue exposure. The calculated RBE values are shown in Figure 1 as functions of the $\alpha_\gamma/\beta_\gamma$ ratio. It shows that for early-responding tissues the RBE value may exceed 1.2. This is significantly higher than the previously reported RBE of 1.10 ± 0.01.[5] Figure 1 also suggests that the RBE value for late-responding tissues can be even higher, especially at low doses. This latter observation agrees qualitatively with data from the study by Gerweck and Kozin.[7] However, the results for late-responding tissues should be viewed with caution because the assumed RBE value of 1.1 at 12 Gy was derived from experimental data that was mostly for early-responding tissues. Furthermore, the uncertainties of the data in Figure 1 are rather large. This is demonstrated by the error bars shown for three data points on the 30-Gy curve. The confidence intervals were calculated with the assumption that the confidence interval for the RBE value measured at 12 Gy per fraction was 1.0-1.2, and Eq. (2) was used for error propagation. It can be seen that the uncertainties in RBE value are much larger than the previously reported[5] standard deviation of 0.01 suggests.

4. Conclusion

The above results were derived using a method based on the linear quadratic model and that was tested using prior experimental data. The results suggest that there is a risk of significant deviations from the widely accepted RBE value of 1.1. This study reports the estimated magnitude of such deviations. The results also indicate that an RBE value of 1.1 is likely to be an underestimation especially for late-responding tissues receiving low proton doses. Therefore, it is important for future studies to provide more RBE data for clinically relevant fractionation regimens for a broad range of tissue types.

Conflict of interest

The authors declare that they have no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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