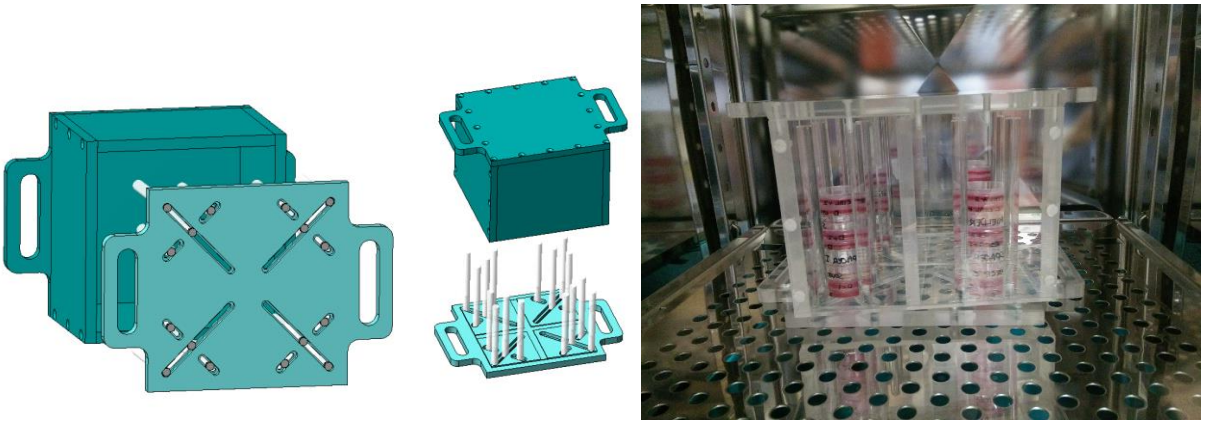
Supplemental Material

Design of experimental container

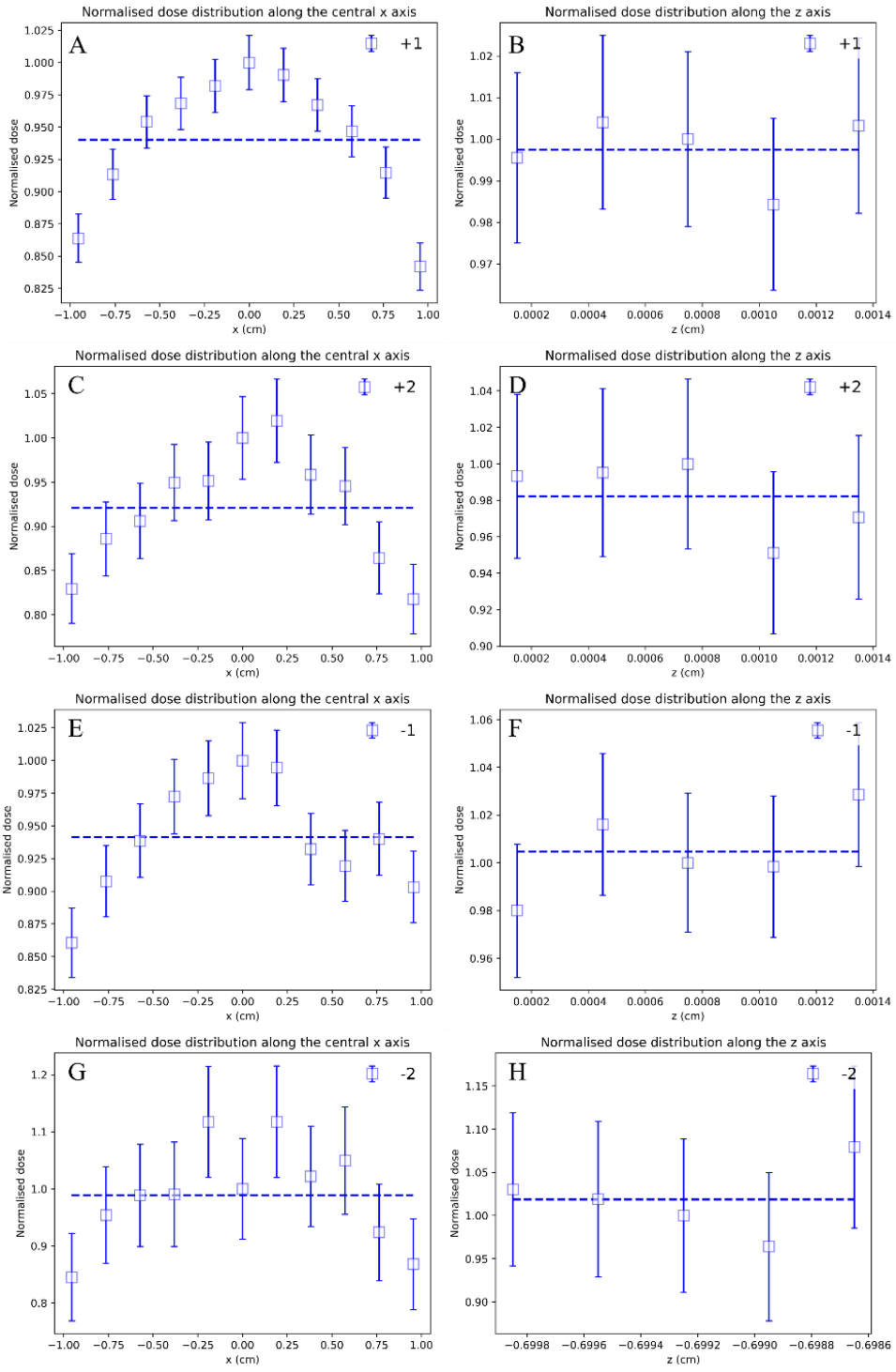
A specially constructed container was used to safely hold the radioactive stacks of dishes. The container made of clear 1 cm thick Perspex to fully attenuate ^{90}Y β^--particles while minimising the creation of bremsstrahlung radiation, provided safety and shielding for radiation workers (Supplemental Figure 1). Moveable posts were constructed to accommodate stacks of variable diameter cell culture dishes. The screws for holding the posts and container together were made of nylon.

A **B**



SUPPLEMENTAL FIGURE 1. (A) A schematic of the Perspex experimental container (stack-separating walls not shown). The container can fit up to four stacks of dishes which can be fixed in place by movable posts and it can also accommodate a single stack of larger dishes if the two perpendicular plastic walls in the centre are removed. (B) Set-up for clonogenic survival showing four stacks of cell-containing dishes inside an incubator.

Dose distribution over irradiated cells in treatment dish

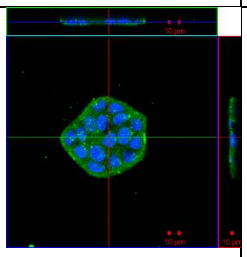
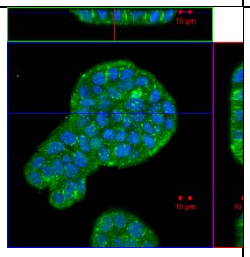
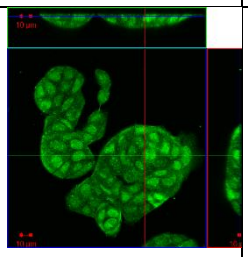


SUPPLEMENTAL FIGURE 2. Normalised simulated-dose distributions along the central x- and z-axes of the 15-µm thick monolayer in dish "+1" (A and B), "+2" (C and D), "-1" (E and F) and "-2" (G and H)

described in Figure 1. Each dose bin in A, C, E and G has a size of 0.19 mm × 0.19 mm × 15 μm and each dose bin in B, D, F and H has a size of 2.1 mm × 2.1 mm × 3 μm. The value of each dose bin is normalised to the value of the dose bin at the centre. Error bars represent standard deviations. A dashed line drawn in each figure represents the mean dose across the monolayer. The deviation is approximately 5% and 2% from the mean dose for dose bins in central x- and z-axes, respectively. All simulations were performed according to the method described in “Monte Carlo (MC) modelling” in the Materials and Methods.

Determination of cell height by confocal imaging

SUPPLEMENTAL TABLE 1. Four HT-29 cell samples were stained and imaged using confocal microscopy. Cell height was derived by estimating the average distance between the culture dish surface and the top of the cells as viewed from cross section along the 3D z-stack confocal images. Area was measured using ROIs in Fiji/ImageJ software. Individual cells were counted manually.

Sample	**1**	**2**	**3**	**4**
Cell line	HT29	HT29	HT29	HT29
Cells plated	3,000	10,000	50,000	10,000
Colony formation	3 days	3 days	3 days	3 days
Colony area	5,500 μm^2	13,000 μm^2	17,000 μm^2	-
Cell count	16	≈50	≈70	≈80% confluent
Cell area (area/count)	340 μm^2/cell	260 μm^2/cell	240 μm^2/cell	-
Cell height	10 µm	16 µm	15 µm	19 µm
Cell volume (area×height)	3,400 μm^3/cell	4,160 μm^3/cell	3,600 μm^3/cell	-
Image				Not collected
Stain	Actin (green) and DNA (DAPI, blue)	Actin (green) and DNA (DAPI, blue)	Actin (green) only	Actin (green) and DNA (DAPI, blue)

Derivation of Equation 1

Using the MIRD formulism, dose (in Gy) was calculated as a function of cumulated activity and the dose per unit cumulated activity (*S*-value). For the experimental setup used here, the time-activity curve follows exponential decay

$$A(t) = A_0 e^{-\lambda t} \tag{S1}$$

where the activity at time t is $A(t)$, the initial activity is A_0, and λ is the physical decay constant given by $\lambda = \ln 2 / T_{phys}$ with T_{phys} the half-life of ^{90}Y.

As the continuous evaporation of media in the apparatus was variable and significantly affected the depth of attenuating matter that the beta radiation traversed, Monte Carlo modelling was performed to derive *S*-values using the measured initial and final media volumes i.e. $S_{initial}$ and S_{final}. A linear function was used to describe the evaporation, such that the rate of change $\left(\frac{S_{final} - S_{initial}}{T}\right)$ during the total exposure period (T) is the slope (m) and the intercept (b) is the initial *S*-value ($S_{initial}$). This slope is expected to remain fixed for each stack as the evaporation was assumed to be at a constant rate since the cylindrical geometry of the culture dishes means that the surface area is constant.

$$S(t) = mt + b = \left(\frac{S_{final} - S_{initial}}{T}\right) t + S_{initial} \tag{S2}$$

Combining the time-activity curve and linear function of $S(t)$ result in the final dosimetry equation where $D(T)$ is the dose for cells exposed to ^{90}Y for duration T:

$$\begin{aligned} D(T) &= \int_0^T \dot{D}(t)\, dt \\ &= \int_0^T A(t) S(t)\, dt \\ &= \int_0^T \left(A_0 e^{-\lambda t}\right)(mt + b)\, dt \end{aligned}$$

$$= \int_0^T mtA_0e^{-\lambda t} + bA_0e^{-\lambda t}\, dt$$

$$= A_0m\int_0^T te^{-\lambda t}\, dt + A_0b\int_0^T e^{-\lambda t}\, dt$$

$$= \frac{A_0m}{\lambda^2}\left[1 - (1+\lambda T)e^{-\lambda T}\right] + \frac{A_0b}{\lambda}\left[1 - e^{-\lambda T}\right]$$

$$= \frac{A_0}{\lambda}\left[\frac{m}{\lambda}\left(1-(1+\lambda T)e^{-\lambda T}\right) + b\left(1-e^{-\lambda T}\right)\right]$$

$$= \frac{A_0}{\lambda}\left[\frac{S_{final}-S_{initial}}{\lambda T}\left(1-(1+\lambda T)e^{-\lambda T}\right) + S_{initial}\left(1-e^{-\lambda T}\right)\right] \qquad \text{(S3)}$$

Relationship between the Lea-Catcheside factor from 6-day and indefinite exposures

The Lea-Catcheside factor, G of an exposure of duration T, for an exponentially decaying dose-rate can be extracted from Roger Dale's paper on dose-rate effects in targeted radiotherapy (Dale, 1996):

$$G(T) = G_T = \frac{\lambda^2}{(\lambda-\mu)\left(1-e^{-\lambda T}\right)^2}\left[\frac{2\left(1-e^{-(\lambda+\mu)T}\right)}{\lambda+\mu} + \frac{e^{-2\lambda T}-1}{\lambda}\right] \quad \text{(S4)}$$

where $\lambda = \frac{\ln 2}{T_{\text{phys}}}$ is the radionuclide decay constant and $\mu = \frac{\ln 2}{T_{\text{rep}}}$ is the damage repair time constant. When $T \to \infty$, $G(T)$ is reduced to Equation 4 in the main text. All clonogenic experiments were performed for cells exposed to ^{90}Y β^--particles for 6 days, therefore the quadratic terms from the SF curves were effectively $G(T = 6\ days) = G_6$ instead of the Lea-Catcheside factor of indefinite exposure, $G(T \to \infty) = G_\infty$.

Derivation of EQD as a function of D_{90Y}

Using the general LQM formalism, for a particular biological response, the negative natural logarithm of the SF, or "log cell kill", E for EBRT relates the dose d in Gy given in n fractions as

$$E_{\mathrm{EBRT}} = -\ln(SF_{\mathrm{EBRT}}) = nd\alpha_{\mathrm{EBRT}}\left(1 + \frac{d}{(\alpha/\beta)_{\mathrm{EBRT}}}\right)$$

where nd in the equation above is the equivalent dose required in a particular fractionation scheme to achieve a specific biological effect, i.e. the EQD. Similarly, for ^{90}Y SIRT with cumulative physical dose D_{90Y}, biological response is given by

$$E_{90\mathrm{Y}} = -\ln(SF_{90Y}) = \alpha_{\mathrm{EBRT}} D_{90Y}\left(RBE_{max} + \frac{G_{\infty} D_{90\mathrm{Y}}}{(\alpha/\beta)_{\mathrm{EBRT}}}\right)$$

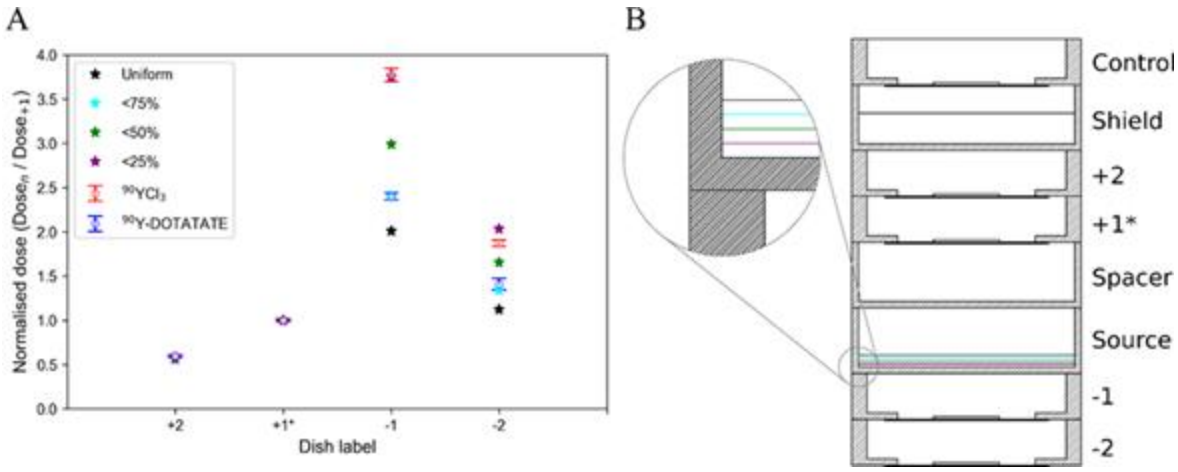
where $RBE_{max} = \alpha_{90Y}/\alpha_{EBRT}$ captures the difference in the linear term $\alpha_{90\mathrm{Y}}$ from that of α_{EBRT} due to differing LET. Equating the biological response in the equations above as follows, yields the iso-effect relationship between EBRT and ^{90}Y SIRT.

$$E_{\mathrm{EBRT}} = E_{90\mathrm{Y}}$$

$$nd\alpha_{\mathrm{EBRT}}\left(1 + \frac{d}{(\alpha/\beta)_{\mathrm{EBRT}}}\right) = \alpha_{\mathrm{EBRT}} D_{90Y}\left(RBE_{max} + \frac{G_{\infty} D_{90\mathrm{Y}}}{(\alpha/\beta)_{\mathrm{EBRT}}}\right)$$

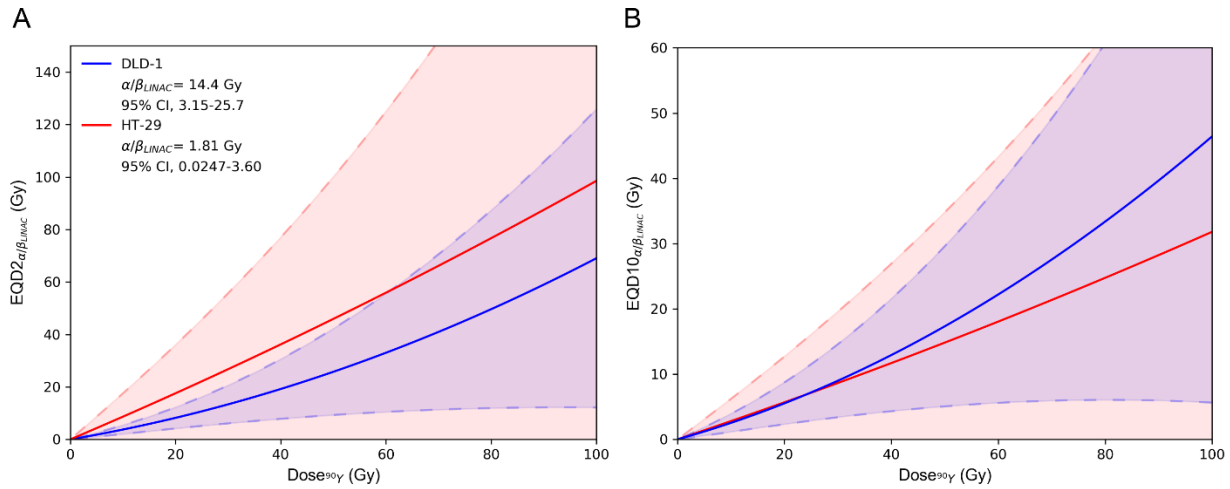
$$EQD_{(\alpha/\beta)_{EBRT}} = nd = \frac{D_{90\mathrm{Y}} \cdot \left(RBE_{max} + \frac{G_{\infty} D_{90\mathrm{Y}}}{(\alpha/\beta)_{\mathrm{EBRT}}}\right)}{\left(1 + \frac{d}{(\alpha/\beta)_{\mathrm{EBRT}}}\right)} \quad \text{(S5)}$$

Since $EQD_{(\alpha/\beta)_{EBRT}}$ is a function of $D_{90\mathrm{Y}}$, one can now relate the physical dose of ^{90}Y SIRT to an equivalent dose of EBRT in terms of biologically equivalent cell kill.



SUPPLEMENTAL FIGURE 3. Comparison of the relative dose distribution in "dry" conditions determined using EBT3 film with MC simulated dose. Dose in each dish was normalised to the dose in stack position +1 (marked with *). Simulated dose distributions (star-shaped) for hypothetical ^{90}Y concentration gradients are shown (purple for <25%, green for <50%, cyan for <75%, and black for a uniform distribution). Measured distributions for $^{90}YCl_3$ (red) and ^{90}Y-DOTATATE (blue) were closely mimicked by simulated distributions for <25% and <75% of source medium, respectively. Error bars represent the standard deviations calculated from the two experiments for each type of radiopharmaceuticals. (B) Schematic of the stack geometry used for calibration. In the expanded circular view, coloured horizontal lines represent different ^{90}Y concentration gradients used in the MC simulations.

$EQD2$ and $EQD10$ as a function of ^{90}Y physical dose, D_{90Y}



SUPPLEMENTAL FIGURE 4. (A) $EQD2$ and (B) $EQD10$ are plotted as functions of ^{90}Y total absorbed dose for DLD-1 (blue) and HT-29 (red) cell lines. The shaded area represents the 95% confidence interval (CI).

REFERENCES

Dale R G 1996 Dose-rate effects in targeted radiotherapy *Phys Med Biol* **41** 1871

Dale R G and Jones B 1999 The assessment of RBE effects using the concept of biologically effective dose *Int J Radiat Oncol Biol Phys* **43** 639-45