Radiolabeled Antibody Combined with External Radiotherapy for the Treatment of Head and Neck Cancer: Reconstruction of a Theoretical Phantom of the Larynx for Radiation Dose Calculation to Local Tissues

Anthony M. Maraveyas, Melvyn Myers, Nick Stafford, Gail Rowlinson-Busza, J. Simon W. Stewart, and Agamemnon A. Epenetos

ABSTRACT

We propose to use radiolabeled antibodies in combination with external beam radiotherapy to improve locoregional control of head and neck cancer. In this case radiation toxicity to mucosa may become a dose-limiting factor and a calculation of the possible compensatory decrease to the external beam radiotherapy would be needed. For this purpose, the following theoretical phantom of a representative organ of this anatomic region, the larynx, was reconstructed and local dosimetric data were derived for a selection of β-emitting isotopes.

The phantom was reconstructed as cylindrical concentric tubes using the established values of an outer diameter of 38 mm and a height of 44 mm. Published mean adult larynx weight (28g) and cartilage weight (14.7 g) were used. Mean mucosa weight from 5 mucosa samples of our patients was calculated to be 2.0 ± 0.4 (SD) g. The remaining weight was apportioned to a fat/muscle compartment (11.3 g). The specific gravity of cartilage (1.10 g/cm3), mucosa (1.04 g/cm3), and fat/muscle (1.04 g/cm3) were used to cross-check the volume/mass disparity of the theoretical tubular tissue shells. The established maximum glottic diameter of 24 mm was used to calculate the central air column volume. Mean laryngeal tumor volume from 8 representative laryngeal tumors was 4.4 ± 3.1 cm3. Tissue compartment thickness was 660 pm for mucosa, 3330 pm for muscle/fat, and 3320 pm for cartilage. These values allowed the calculation of dose absorbed fractions for a number of theoretical radioimmunoconjugates by extending the established calculation of absorbed fractions for spheres of known diameter to absorbed fractions of tissue planes (annuli) of known thickness. We calculated a D∞ for the respective tissues in the larynx for 131I-, 185Re-, 18Re-, 65Cu-, 111In-, and 153Sm-labeled HMFG1. Compensatory decrease to the external radiotherapy dose is 1.1 Gy for each injection of the radioimmunoconjugate we propose to use (131I-HMFG1). This would be best implemented through the modification of the external radiotherapy fractions falling within 2 effective half-lives of this radioimmunoconjugate in the mucosa.

INTRODUCTION

Radiolabeled antibodies are currently being assessed for the possibility of treating a variety of malignancies. They are generally viewed as a systemic form of therapy. It has emerged, however, from the pharmacokinetic and biodistribution profile of the systemically administered intact murine IgG molecule, that it is a rather ineffective radiation vehicle (1). The amounts of radioactivity reaching the tumor are compatible with very low radiation doses, usually on the order of 1–15 Gy depending on isotope, absolute uptake, residence time, and number of administrations. With the exception possibly of lymphomas (2), durable responses in bulky solid tumors have been rare. Methods of improving the technology as a systemic treatment of cancer are being continuously investigated. Bifunctional antibodies (3), enzyme-prodrug activation strategies (4), regional administration (5), improved chelating agents (6), and intratumor administration (7) are a small selection of the suggested strategies to directly or indirectly ameliorate dosimetry.

We have proposed that the currently attainable small radiation dose may still be used to a beneficial effect to improve locoregional control as a supplementary dose to conventional external beam radiotherapy (8).

Radiosensitive tumors treated with a curative intent and for which local control is of vital importance for increased disease-free survival seem the best candidates for this strategy. Head and neck squamous cell carcinoma is typical of such a tumor. Increased local control may lead to longer survival. The idea is that a bimodal radiation treatment, not unlike the classically used implant brachytherapy and external beam radiotherapy (9), may be a feasible way of extracting maximum gain from the technology as it currently stands. If the steep dose-response curve that generally characterizes these tumors continues above the currently admissible doses of 65–70 Gy, even a small radiation increase that spares normal structures may confer a relatively large benefit (10).

Bone marrow depression, which is the major dose-limiting toxicity of radioimmunotherapy, is reached long before any other body tissue manifests any radiation effect. Stewart et al. (11, 12), for example, using peritoneally implanted LiF thermoluminescent dosimeters demonstrated the relatively low radiation dose delivered to the peritoneal cavity lining even by a regionally administered 131I- or 185Re-radioimmunoconjugate, as compared to a radioactive colloid. In our case, however, a detailed study of the radiation dose to normal tissues is warranted, because external beam radiotherapy is limited by acute local tissue reaction, usually mucosa, and the fear of long term radiation effects (radiocarcinosis) on other structures. Increased adverse effects could be prevented through a detailed knowledge of the dose delivered to each of the local structures by the antibody. Hence a compensatory reduction to the external beam can be calculated. It is with this goal in mind that we undertook the reconstruction of a phantom of the larynx, a representative anatomical structure of the "head and neck." To illustrate the modification of the actual dosimetry that the application of the phantom generates, depending on individual tissue volume and β-emitting isotope studied, we have used as an example biodistribution and pharmacokinetic data derived from our study of patients with head and neck carcinoma with the mAb HMFG1.2

MATERIALS AND METHODS

mAb HMFG1. This antibody, originally designated 1.10.F3, was raised against the HMFG. HMFG1 (IgG1) was developed from a mouse receiving a further boost of HMFG and recognizes an antigenic determinant on the @re protein of the HMFG designated polymorphic epitheial mucin of a high molecular weight (M, > 400000) composed of tandem repeats of a proline-aspartic acid-threonine-arginine-proline motif (13). Variation in glycosylation

Received 7/11/94; accepted 1/3/95.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked 'advertisement' in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

1 A. M. acknowledges the financial support of the A. G. Leventis Foundation. To whom requests for reprints should be addressed.

2 The abbreviations used are: HMFG, human milk fat globules; AUC, area under the curve; ER, external radiotherapy; MIRD, medical internal radiation dose; i.e., intrathcal; RIT, radioimmunotherapy.
patterns of polymorphic epithelial mucin are proposed to explain the difference in antigen positivity patterns noted for a variety of adenocarcinomas. This antibody reacts strongly with a wide range of human adenocarcinomas and squamous cell carcinomas (14). Radiolabeled HMFG1 has been used in many clinical studies for the diagnosis and treatment of human malignancies (11, 12).

**Radioiodination with 125I.** The one vial Iodo-Gen method was used (15). Briefly, 1,3,4,6-tetrachloro-3a,6a-diphenylglycoril (Iodo-Gen; Pierce, Chester, United Kingdom) was used for the covalent linking of HMFG1 to 125I (Amersham International, Amersham, Buckinghamshire, United Kingdom). A chloroform solution containing 5 mg/ml of Iodo-Gen was made. Aliquots of 100 μl (50 μg) in sterile NUNC tubes were left overnight in a fume hood to evaporate. These were subsequently stored at −20°C and used as required. The pH of the protein solution was adjusted to 7.5 using 1 M Tris, pH 9.5. Protein was mixed with 125I in a tube containing 50 μg Iodo-Gen at room temperature, rotating for 10 min. Free iodine was separated from labeled protein using a 20-ml Sephadex G-50 column (Pharmacia, Uppsala, Sweden). The column was eluted with PBS. Fractions of 2 ml were collected and the activity of each fraction was counted in a gamma counter. The fractions containing the protein peak were collected and filtered through a 0.22 μm Millipore filter (Millipore, Watford, United Kingdom). Labeling efficiency was calculated by the ratio protein bound activity/activity loaded on the column and specific activity by the value of protein bound activity (mcI/μg protein). The purity and the stability of the conjugates were tested by fast protein liquid chromatography. Radioimmunoactivity was assessed in a semiquantitative method by solid phase RIA on purified antigen coupled to BSA (16). All patients were given injections, within 3 h of labeling, of 160 μCi 125I-HMFG1.

**Patients.** The study has been approved by the Ethics Committee of the Hammersmith Hospital. Written informed consent was obtained from all patients entered in the study. All patients without a history of major allergic reactions to proteins or prior injection of mAbs and undergoing surgery for primary squamous cell carcinoma of the head and neck were eligible for the study.

Fourteen patients with primary head and neck cancer [nine males and five females, mean age, 61 ± 9 (SD) years] were given injections of HMFG1 antibody. The anatomical locations of the tumors are presented in Table 1.

**Protocol of Biodistribution Study.** Patients were given i.v. injections, at fixed time points (24–48 h or 72 h), prior to surgery, of 125I-labeled HMFG1 (90–125 μg/160 μCi). Immediate blood samples (5–6 samples in the first 30 min) after injection were drawn. A 3–4-ml blood sample at 1–2 h and a daily sample for the following 4 days were obtained. A blood sample was drawn at the time of the surgical excision of the tumor. Samples were clearly and accurately labeled with the time and date of venesection. A 24-h urine collection was initiated immediately in order to collect the total urine excreted following injection and was maintained for 72 h. At surgery tissue samples were obtained. These included tumor tissue from the primary site and tumor-invaded distal sites (e.g., lymph nodes) where available. Specimen size was at least 8 x 4 x 4 mm. No panendoscopy biopsies were included in these studies. Samples of normal tissues (skin, muscle, fat, cartilage, lymph node, tonsil, etc.) were also obtained when and if available. Areas of normal tissues ailed by electrocautery were avoided. All tumor samples were bisected; one half was fixed and the other half was stored in liquid nitrogen. Normal tissue samples were fixed. Standards of the injection material were made; the tissue, blood, and urine samples were weighed and then measured for activity together with the injected dose per kg of tissue.

**Method for Measuring Mucosa Surface Area/Weight.** An adaptation of the method of Stell et al. (17–19) was used. The surgical specimens of mucosa were fixed in formol-saline for at least 2 days. They were then blotted dry and spread out on a thin polystyrene sheet. A second polystyrene sheet was used to cover the tissue and the sheets were subsequently pressed together using bulldog clips. A thin black felt pen was used to outline the mucosal surface on the polystyrene sheet and then 2 cutouts of each mucosa specimen were made by two different observers on A4 Daler cartridge paper pad. Standards of known surface area were made from this paper and plotted as a function of their weight to obtain a standard curve. Each time a new piece of paper was used new standards were made. The weights of the mucosa cutouts were determined and a surface area was assigned to them by solving the equation of the standard curve for the unknown surface variable (Fig. 1). Five mucosa samples from four patients were thus studied. Three samples of laryngeal mucosa were from total laryngectomy specimens and a further two samples (one of the laryngeal mucosa and one from the esophageal region) were from the same patient who underwent pharyngolaryngoesophagectomy and stomach pull-up for a pyriform fossa tumor. Table 3 presents the individual measurements.

**Data for Reconstruction of the Phantom.** Three dimensional information and information on the weights of the larynx and cartilages was obtained from the following established values. Volume of the larynx for adult male (20): length, 44 mm; transverse diameter, 43 mm; sagittal diameter, 36 mm; mean diameter, 38 mm. Cartilage mass for adult male (21): thyroid cartilage, 8.6 g; cricoid cartilage, 5.3 g; arytenoid cartilage, 0.39 g X 2; total cartilage weight, 14.7 g. Mass of whole male adult larynx (22): 28 g. Mass of the corresponding mucosa lining was indirectly calculated as described using the following information. Mucosa surface area for adult male: (a) glottic surface area (17), 70.5 ± 3.45 mm2; (b) subglottic space surface area (18), 1297.5 ± 36.7 mm2; (c) supraglottic space surface area (20), 2202.4 ± 74.71 mm2; (d) total laryngeal mucosal surface area, 35.7 cm2. Mass of laryngeal musculature/adipose tissue compartment: this was calculated by subtracting the sum of the known cartilage (Cc) and mucosa mass (Mc) from the total mass of the larynx (Lm):

\[ L_m - (C_c + M_c) \]

(A)

Adipose tissue comprises a small proportion of the weight essentially concentrated in the space between the thyroid cartilage and the anterior laryngeal wall. From our biodistribution data there is no significant difference in uptake between muscle and fat although clearance is more prolonged for muscle. We chose to apportion the remaining tissue weight as a muscle/adipose tissue compartment to derive a relative volume and used the muscle clearance data for dosimetry, therefore sustaining a small overestimate of radiation dose to this tissue.

---

**Table 1 Anatomical site of tumors in the patients studied**

<table>
<thead>
<tr>
<th>Anatomical site</th>
<th>No. of patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Larynx</td>
<td>7</td>
</tr>
<tr>
<td>Tongue</td>
<td>2</td>
</tr>
<tr>
<td>Pyriform fossa</td>
<td>3</td>
</tr>
<tr>
<td>Skin</td>
<td>2</td>
</tr>
</tbody>
</table>
Calculations for the Relative Masses and Volumes. From the established laryngeal dimensions \((20)\) a mean diameter of 38 mm for the phantom was used. The established physiological length of the larynx of 44 mm was used. Using the equation

\[
V = \pi \cdot r^2 \cdot h
\]

the volume of the cylinder which represents the total laryngeal volume \((V_c)\) was calculated.

An air column volume \((V_{AC})\) was calculated using the maximum established diameter of the glottic space \((23)\) which is 2.4 cm. This was subtracted from the whole volume of the laryngeal cylinder leaving a total tissue volume \((V_{RT})\). Individual tissue volumes were apportioned based on the relative masses, i.e. mucosa volume

\[
V_{MC} = \frac{11.3}{28} \cdot V_{RT}
\]

muscle/adipose volume

\[
V_{MU} = \frac{2/28}{2} \cdot V_{RT}
\]

cartilage volume

\[
V_c = \frac{14.7}{28} \cdot V_{RT}
\]

The relative position of each tissue in the phantom was allocated as mucosa on the inner cylinder shell (annulus) followed by muscle/fat and the outer shell consisting of the cartilage.

The following methodology of calculating the relative cylinder diameter was followed. First a radius \((r_3)\) for a cylinder \((V_{C3})\) comprising the dead space and the volume of the mucosa was calculated; then the thickness of the shell \((\theta_3)\) was found by subtracting the radius of this cylinder from the radius of the dead space cylinder \((r_1)\). The same calculations were made for the muscle/adipose cylinder volume \((V_{C2})\) from which a radius \((r_2)\) was obtained and by subtracting \(r_3 - (r_2 + r_1)\) the thickness \((\theta_2)\) of the muscle compartment was obtained. Similarly a radius \((r_4)\) for the cartilage cylinder volume \((V_{C1})\) which coincides with \(V_{C2}\) was obtained and the thickness of the cartilage compartment \((\theta_1)\) was derived from the equation \(r_4 - (r_3 + r_2 + r_1)\).

**Tumor Dimensions.** The tumor dimensions and volume in the eight laryngeal cancer specimens were calculated as

\[
T_c = \frac{4}{3} \cdot \pi \cdot H \cdot W \cdot L
\]

where \(T_c\) is the tumor volume, \(H\) is height, \(W\) is width, and \(L\) is length of the tumor as measured on the pathological specimen.

**Specific Gravity of Relative Tissues \((23)\).** The specific gravity of muscle is 1.040 g/cm\(^3\), that of mucosa is 1.040 g/cm\(^3\), that of fat is 0.95 g/cm\(^3\), and that of cartilage is 1.10 g/cm\(^3\). These values were used to check the discrepancy between the theoretical tissue volume represented in the phantom and the actual volume of the mass of the tissues included.

A linear correlation of mass and volume for a xenograft derived from a squamous cell carcinoma cell line (H.Ep-2) was used in lieu of specific gravity to calculate the mean mass of the human tumors.\(^3\)

**Dosimetry Calculations.** In this study dosimetric calculations, based on the MIRD formulations \((24)\) for internally administered isotopes, were performed only for the local normal structures (muscle, cartilage, and mucosa) and for the tumor. Whole body dosimetry based on the acquired data is not the subject of this study \((25)\). Furthermore absorbed dose contribution to the above structures from the remainder of the body or from proximal tissues which may accrue substantial activity (i.e., the thyroid for \(^{131}\)I-based treatments) are not included in the calculations. The calculations were carried out for a selection of \(\beta\)-emitting isotopes that have at times been proposed as radioimmunoconjugates. Isotope activity curve was derived from the areas corresponding to the first 3 days after decay correction plus the assumed area from day 3 to infinity.

To calculate the accumulated activity after the three studied time points \((A_{3 \to \infty})\), exponential decay was assumed using the formula

\[
A_{3 \to \infty} = t_{1/2 \alpha} \cdot 1.44 \cdot A_3
\]

where \(A_3\) represents antibody uptake (i.e., decay corrected) at 3 days. The effective half-life \((t_{1/2 \alpha})\) was calculated as

\[
\frac{1}{t_{1/2 \alpha}} = \frac{1}{t_{1/2 \omega}} + \frac{1}{t_{1/2 \nu}}
\]

Antibody half-life \((t_{1/2 \omega})\) was calculated for each tissue separately after fitting an exponential curve to the three existing time points.

The absorbed dose \((D)\) for 1 g of tissue uniformly irradiated by the isotope is given by the MIRD equation:

\[
D_\omega = A_{0 \to \omega} \cdot \Sigma D_\omega \cdot \phi
\]

where \(D_\omega\) is the equilibrium dose constant for the isotope studied in g . Gy/μCi. For particles \(i\) emitted in the target for a particular type and energy.

**Derivation of the Absorbed Fractions.** Data on the mean absorbed fraction for each isotope for a volume distribution in spheres of known radius were available \((26)\). These were modified to calculate the absorbed fraction for the particular geometry of each organ using numerical integration of volumes. The geometries were cylindrical shells (annuli) for cartilage, muscle, and mucosa and an oblate spheroid for the tumor \((2f)\).

Each cylindrical shell of thickness, \(\delta\), can be thought of as a layer of contiguous activity filled spheres of radius \(\theta/2\). If the absorbed fraction for each sphere is \(\phi\) then the fraction of particles escaping from the sphere is \((1 - \phi)\). Of those escaping about one-half will be absorbed along the length of the cylindrical shell. The total fraction absorbed is thus

\[
\phi + \frac{(1 - \phi)}{2}
\]

Values of \(\phi\) for small spheres filled with the appropriate radionuclides were obtained from Bardies and Chatal \((26)\). Absorbed fractions were equal or close to unity for the larger organs and tumor and for lower \(\beta\) emitters such as \(^{131}\)I but significantly less for the thin mucosa and the higher \(\beta\) energies of \(^{90}\)Y and \(^{186}\)Re.
Table 3: Surface area and corresponding weight of the available mucosa samples

Calculation was made by the cutout method described. By extrapolating the derived values for the mass of the tissue to the established surface area by Stell et al. (17-19), the values in the last column were determined. A value of 55 µg/cm² of mucosa was derived.

<table>
<thead>
<tr>
<th>Patient/specimen</th>
<th>Surface area (cm²)</th>
<th>Specimen mass (g)</th>
<th>Mucosa mass (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>E. K./larynx</td>
<td>1.345</td>
<td>0.09</td>
<td>2.40</td>
</tr>
<tr>
<td>R. F./larynx</td>
<td>1.42</td>
<td>0.08</td>
<td>2.00</td>
</tr>
<tr>
<td>P. M./larynx</td>
<td>2.84</td>
<td>0.12</td>
<td>1.50</td>
</tr>
<tr>
<td>A. vD/larynx</td>
<td>1.67</td>
<td>0.11</td>
<td>2.35</td>
</tr>
<tr>
<td>A. vD/pharyngoosophageal</td>
<td>3.18</td>
<td>0.15</td>
<td>1.70</td>
</tr>
</tbody>
</table>

RESULTS

Mean Mucosa Mass. Table 3 depicts the individual mucosa weights and corresponding mucosa surface of the five samples studied. Mean mucosa mass was found to be 2.0 ± 0.4 g or 55 µg/cm².

Tumor Dimensions. Table 4 gives the individual measurements for each of the specimens. Mean tumor depth (D) was 1.3 ± 0.3 cm, mean tumor width (W) was 2.2 ± 0.6 cm, and mean tumor length (L) was 2.9 ± 0.8 cm. The mean tumor volume was calculated to be 4.4 ± 3.1 cm³.

Mass of Laryngeal Musculature/Adipose Tissue Compartment. The mean mass for this compartment was calculated to be 11.3 g.

Values for the Relative Masses and Volumes. Laryngeal volume (V_L), 50 cm³; air column volume (V_Ac), 20 cm³; total tissue volume (V_TB), 30 cm³; mucosa volume (V_Mu), 2.14 cm³; muscle/adipose volume (V_Ma), 12.1 cm³; cartilage volume (V_C), 15.75 cm³.

Values for the Cylinder Radii and Compartment Thickness. r_1 = 1.2 cm; r_2 = 1.266 cm; r_3 = 1.57 cm; r_4 = 1.9 cm; θ_1 = r_2 - r_1 = 0.066 cm; θ_2 = r_3 - (r_2 + r_4) = 0.31 cm; θ_3 = r_4 - (r_3 + r_2 + r_4) = 0.33 cm. These measurements were used to apportion the volumes in the illustrated phantom annuli (Fig. 2A) and subsequently to calculate the absorbed fractions (Table 5). Consistency Cross-Checking Calculation. The mean laryngeal measurements derived from the various sources as stated in “Materials and Methods” led to a tissue volume of 30 cm³. Using the respective specific gravities (23) the actual tissue mass of 28 g would have corresponded to a tissue volume of 26.2 cm³.

The mucosa surface (S_m) of the phantom was derived from

\[ S_m = \pi \cdot D \cdot h \]

where h is the height of the cylinder and D is the diameter of the air column volume; this leads to a value of 33.2 cm², a value again close to the 35.7 cm² as measured by Stell et al. (17-19) for actual specimens. Taking into account that the mass of the larynx is also made up of an unknown proportion of adipose tissue with a specific gravity of 0.9 g/cm³, one appreciates that the discrepancies in both measurements are relatively small. The parameters therefore of the theoretical model, despite the use of data obtained from a variety of sources and an indirect methodology of calculation, are quite consistent.

Antibody Residence Time in Tissues. Fig. 3 depicts the clearance of ¹²⁵I-HMFG1 from the normal tissues and the tumor. Complete pharmacokinetic and biodistribution data have been obtained for muscle, mucosa, adipose tissue, and tumor (25). For cartilage only, uptake at day 1 was measured in three different samples from the superior cornu of the thyroid cartilages. A mean value of 0.6% injected dose/kg was obtained. A single sample of auricular cartilage and thyroidal cartilage (superior cornu) was obtained for days 2 and 3, respectively. No mean value could therefore be derived. However, because no antibody cross-reactivity with the cartilage exists (immunostaining data not shown), the vascularity of the cartilage would be, if anything, less than that of adipose tissue, and the small number of values obtained had no significant difference from those of adipose tissue, the exponential clearance and AUC₁⁻₃ for adipose tissue was used as a guideline to derive values for cartilage. Because the initial uptake in adipose tissue was 0.8% injected dose/kg, a possible small overestimate of the dose to cartilage may result.

The biological half-lives used for the calculation of the effective half-lives, in turn used to derive the AUC₃ → ∞ for each tissue, were obtained from fitting an exponential curve to the existing time points.
Exponential curves (Fig. 4) were, respectively: mucosa, 62 h; muscle, 123 h. The antibody half-life in the tumor has been calculated as 123 h.

DISCUSSION

The current use, for immunotherapeutic purposes, of a number of β-emitting agents and the further assessment of other promising candidates has led to the need for better understanding of the radiation effects of internally administered isotopes. Despite progress in the field the only practicable approach in the clinic of computing dosimetry for internally administered radiobiological agents is through the use of the MIRD formalisms. Recently Badger and Fisher (27) have argued in favor of better modeling of the radiation dose and clearer definition of treatment criteria as a means for a more rational understanding of the anticipated dose response from radioimmunotherapy. Because the possibility of local tissue toxicity in this case is increased, two of the most promising routes of administration, the i.p. (28) and the i.t. (29) ones, have been modeled to assess this possibility. Watson et al. (28) described a phantom of the peritoneal cavity designed to calculate dose to the peritoneal lining as a function of penetration depth of a given β-emission from an isotope in solution. Modeling of the peritoneal surface was based on a planar geometrical concept, i.e., a single plane of tissue above which the solution was suspended. Using a similar approach, Millar and Barrett (29) modeled the spinal cord for the i.t. use of radioimmunoconjugates for leukemia. The cord was modeled as two concentric cylinders. The central solid one represented the spinal cord while the concentric annulus represented the cerebrospinal fluid space. Using methodology similar to that of Watson et al. they also calculated dose as a function of depth to spinal cord from the annulus containing the radionuclide (cerebrospinal fluid space).

Table 5. Equilibrium dose constants and calculated absorbed fractions for the normal tissue compartments and tumor for each of the studied isotopes

<table>
<thead>
<tr>
<th>Isotope</th>
<th>$\Sigma_D$ (g rad/μC/rh)</th>
<th>Cartilage ($t_\beta$ 0.33 mm)</th>
<th>Tumor</th>
<th>Muscle ($t_\beta$ 0.31 mm)</th>
<th>Mu cosa ($t_\beta$ 0.07 mm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$^{131}I$</td>
<td>0.40</td>
<td>0.92</td>
<td>1.00</td>
<td>0.91</td>
<td>0.72</td>
</tr>
<tr>
<td>$^{198}Re$</td>
<td>0.76</td>
<td>0.81</td>
<td>1.00</td>
<td>0.80</td>
<td>0.62</td>
</tr>
<tr>
<td>$^{186}Re$</td>
<td>1.70</td>
<td>0.67</td>
<td>0.85</td>
<td>0.65</td>
<td>0.54</td>
</tr>
<tr>
<td>$^{44}Y$</td>
<td>2.00</td>
<td>0.64</td>
<td>0.85</td>
<td>0.62</td>
<td>0.53</td>
</tr>
<tr>
<td>$^{67}Cu$</td>
<td>0.33</td>
<td>0.94</td>
<td>1.00</td>
<td>0.93</td>
<td>0.78</td>
</tr>
<tr>
<td>$^{153}Sm$</td>
<td>0.50</td>
<td>0.90</td>
<td>1.00</td>
<td>0.89</td>
<td>0.72</td>
</tr>
</tbody>
</table>

Fig. 3. Mean time-dependent uptake of the HMFG1 antibody in tumor and the other normal tissues over the three time points studied.

Fig. 4. Exponential curves fitted to the time points of Fig. 3. The derived half-lives are shown.

The more energetic the β-radiation, the greater is the dose diminution caused by the diminishing volume with the effect being more prominent in mucosa. The relatively large mean diameter of tumors as compared to the path length in the tissue of the majority of the isotopes leads to small modifications in dose; however, the absorption profile of dose in smaller tumors or micrometastatic foci from energetic β-particles will be closer to that described for mucosa than for the mean tumor mass (42, 43).
A dose of ~3 cGy/mCi of radioimmunoconjugate injected is expected to be delivered to a tumor of the mean dimensions studied. At the same time the mucosa will be receiving 0.8 cGy/mCi injected. An injected activity of 150 mCi would mean that the mucosa would be receiving an extra 1.1 Gy, the majority over 3–4 days (2 effective half-lives). Muscle and cartilage will be receiving 67 and 48 cGy, respectively. Any modification of the external beam that may be needed to reduce acute local toxicity to mucosa should more than compensate for the small dose to the other normal structures.

Table 6 Dosimetry for the 111In-labeled HMFG1

<table>
<thead>
<tr>
<th>111In-HMFG1 uptake (% i.d./kg)</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
<th>$T_{1/2}$ (days)</th>
<th>$A$ (μCi·h·g⁻¹)</th>
<th>$D$ (cGy/mCi)</th>
<th>T/NT</th>
<th>$\phi$</th>
<th>$D^c$ (cGy/mCi)</th>
<th>T/NT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>5.02</td>
<td>4.24</td>
<td>4.18</td>
<td>3.15</td>
<td>7.3</td>
<td>2.92</td>
<td>1.00</td>
<td>2.92</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mucoa</td>
<td>2.75</td>
<td>2.82</td>
<td>1.42</td>
<td>1.96</td>
<td>2.7</td>
<td>1.10</td>
<td>0.72</td>
<td>0.79</td>
<td>2.65/1</td>
<td>0.72</td>
</tr>
<tr>
<td>Muscle</td>
<td>0.82</td>
<td>0.69</td>
<td>0.53</td>
<td>3.16</td>
<td>1.2</td>
<td>0.50</td>
<td>0.91</td>
<td>0.45</td>
<td>5.8/1</td>
<td>0.91</td>
</tr>
<tr>
<td>Cartilage</td>
<td>0.73</td>
<td>0.47</td>
<td>0.39</td>
<td>2.24</td>
<td>0.7</td>
<td>0.30</td>
<td>0.92</td>
<td>0.28</td>
<td>9.7/1</td>
<td>0.92</td>
</tr>
</tbody>
</table>

$^a$ i.d., injected dose; T/NT, tumor/respective normal tissue ratio.
$^b$ Absorbed fraction.
$^c$ Corrected.

Table 7 Dosimetry for a theoretical 90Y-labeled HMFG1 radioimmunoconjugate

Inability to inject enough of this isotope for a reasonable dose to the tumor has disqualified it as a candidate for the proposed treatment. The improvements in the chelator technology have not led to a substantial increase in injectable activity (33). Furthermore, this isotope will be at a disadvantage when the treatment of coexisting minimal disease in lymph nodes is contemplated, due to similar absorbed fraction considerations as seen for mucosa.

<table>
<thead>
<tr>
<th>90Y-HMFG1 uptake (% i.d./kg)</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
<th>$T_{1/2}$ (days)</th>
<th>$A$ (μCi·h·g⁻¹)</th>
<th>$D$ (cGy/mCi)</th>
<th>T/NT</th>
<th>$\phi$</th>
<th>$D^c$ (cGy/mCi)</th>
<th>T/NT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>4.23</td>
<td>3.00</td>
<td>1.92</td>
<td>1.75</td>
<td>4.5</td>
<td>9.0</td>
<td>0.85</td>
<td>7.65</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mucoa</td>
<td>2.31</td>
<td>1.99</td>
<td>0.85</td>
<td>1.31</td>
<td>1.9</td>
<td>3.8</td>
<td>2.4/1</td>
<td>0.53</td>
<td>2.0</td>
<td>3.8/1</td>
</tr>
<tr>
<td>Muscle</td>
<td>0.69</td>
<td>0.49</td>
<td>0.31</td>
<td>1.76</td>
<td>0.7</td>
<td>1.4</td>
<td>6.4/1</td>
<td>0.62</td>
<td>0.87</td>
<td>8.8/1</td>
</tr>
<tr>
<td>Cartilage</td>
<td>0.61</td>
<td>0.33</td>
<td>0.23</td>
<td>1.43</td>
<td>0.5</td>
<td>1.0</td>
<td>9.0/1</td>
<td>0.64</td>
<td>0.64</td>
<td>11.9/1</td>
</tr>
</tbody>
</table>

$^a$ i.d., injected dose; T/NT, tumor/respective normal tissue ratio.
$^b$ Absorbed fraction.
$^c$ Corrected.

of the larynx being the most sensitive. The clinical peak of symptoms is usually at 2.5–3 weeks. To acquire accurate local dosimetry, which would mean calculating absorbed fractions ($\phi$) for the tissues in question for every isotope candidate, we reconstructed a phantom of a representative organ of the head and neck area: the larynx. This was chosen due to the relative simplicity of the structure and due to the fact that we had precise dosimetric data pertaining to all the major tissues that constitute this organ. The few assumptions (fat/muscle compartment and cartilage time-dependent clearance) we had to make were in such a way that at worst our dose predictions for that normal structure would be slightly higher. Further sources of possible error in our model could stem from: (a) the “spreading technique” we used, modified from the protocol by Stell et al. (17–19). Pressure applied to spread the tissue was through the whole laryngeal mucosa lying on Perspex sheets (the cartilage had been dissected out). We may therefore be slightly underestimating the weight that corresponds to the surface area due to direct pressure applied only through the thickness of the mucosa, causing a more profound spreading effect; (b) the involved samples may have included differing amounts of submucosal tissue which may account for the discrepancies between mucosa specimens. However if one looks at the illustration of the “Swiss-roll” technique (32) of stripping whole mucosas of larynx, one can observe differing thicknesses through the roll that correspond well with the different histological examples of mucosas we have obtained; (c) much of the data, i.e., weight of larynx, corresponding mucosa surface area, volume measurements of larynx and weight of cartilage, were culled from different literature sources.

Mucosa thickness calculation lies within the existing measured examples of nasal and tracheobronchial mucous membranes (23) and, even allowing for inaccuracies, the principle is well illustrated; furthermore, the uptake data we had available were for mucosa samples of the above histological structure (25). It can be assumed that similar dose absorbed fractions will pertain to mucosa of the other normal anatomical structures, e.g., pharynx, esophagus.

The model allows the calculation of dose absorbed fractions for laryngeal mucous membranes. This represents an extension of theoretical sphere models developed to study dosimetry of micro-metastases. Energy absorbed fractions ($\phi$) (Table 5) can now be incorporated in the calculations; hence tumor:normal tissue dose ratios for the different isotopes are modified. To illustrate this point, we have used data from a group of patients in which we have studied the biodistribution and pharmacokinetics of HMFG1 in detail (25). We have shown that while tumor:normal tissue antibody uptake ratios (excluding mucosa) are of the order of >5/1 the tumor/mucosa ratio is only ~2.5/1 due to cross-reactivity of HMFG1 antibody with this tissue. The dosimetric calculations...
Mucosa dose sparing is similar to that of $^{153}$Sm and the same considerations for minimal disease exist. However, dose to tumor is reasonable and the dose rate will be substantially achievable our study goal of a 10–15% dose increment with a putative $^{88}$Re-conjugate.

Based on the MIRD formulae lead to a tumor/mucosa dose advantage ranging from 2.3 to 2.65/1 (Tables 6–11). These values, however, are inaccurate as some of the disintegration energy (up to ~50%) for the more energetic $\beta$-emitters dissipates outside the distribution volume of this tissue. Following incorporation of the absorbed fractions, the corresponding adjusted tumor/mucosa ratios range from 2.85/1 to 3.9/1 with $^{67}$Cu and $^{186}$Re being the extreme examples.

Our whole body and bone marrow dose calculations predict that ~150 mCi of $^{131}$I-labeled HMFG1 can be administered i.v. to a patient as a single bolus (25). This would deliver a dose of ~110 cGy to the mucosa with most of the dose having been delivered in 150 h, i.e., three effective half-lives of the radioimmunoconjugate in this target tissue. The corresponding tumor dose would be about ~440 cGy.

Our dose escalation studies with $^{90}$Y-iodo-octreotide/diethyl-entriaminedepentacetic acid-HMFG1 have led to a maximum admissible dose of 18.5 mCi/m² via the i.p. route (33). Considering the differences in the published pharmacokinetics of the two routes (34) and the respective blood clearance data of the two routes (33) we would expect that ~60% of this dose would be injectable via the i.v. route. This would in effect deliver a ~40 cGy dose to the mucosa but only a ~140 cGy to the mean tumor volume in this area.

Two possible methods of dose adjustment can be discussed; modifying or omitting a single ER fraction to implement the necessary dose reduction or incorporating the dose adjustment over a number of fractions. Because there exists a possibility of using a variety of isotopes, one must consider the dose rate of each radioimmunoconjugate. If fractions of ER are 200 cGy daily or 150 cGy (35) twice daily, which have been associated with maximum mucosa tolerance, then it is our inclination to recommend a daily adjustment of dose individualized for each patient according to the fractionation regimen prescribed, type of isotope, and total activity administered. We envisage no need for adjustment at initial doses of a dose-escalating phase I study. However, as the maximum injectable dose is reached, the fractions falling within two effective half-lives of the radioimmunoconjugate may be the ones needing some modification. If the AUC of the antibody is known, a 24-h dose rate can be easily calculated and the ensuing ER fraction can be modified accordingly. For example, ER fractions over 2–3 days following administration of the $^{131}$I-conjugate may need to be adjusted to take into account the mucosa dose. It is probable that the overall radiobiological effect of the radiotherapy combination as proposed is much more complicated (36–40); however, the lack of pertinent data makes it difficult at this moment to recommend any course of action other than the cautious dose adjustment of ER.

In conclusion we have presented the first phantom of its kind pertaining to dosimetry of internally administered radioactivity for the head and neck. We derived dose absorbed fractions for normal anatomical structures in this

---

### Table 9: Dosimetric calculations for a theoretical $^{186}$Re-HMFG1 radioimmunoconjugate

<table>
<thead>
<tr>
<th>$^{186}$Re-HMFG1 uptake (% i.d./kg)</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>2.0</td>
<td>0.70</td>
<td>0.20</td>
</tr>
<tr>
<td>Mucosa</td>
<td>1.1</td>
<td>0.50</td>
<td>0.10</td>
</tr>
<tr>
<td>Muscle</td>
<td>0.3</td>
<td>0.10</td>
<td>0.04</td>
</tr>
<tr>
<td>Cartilage</td>
<td>0.3</td>
<td>0.08</td>
<td>0.03</td>
</tr>
</tbody>
</table>

---

### Table 10: Dosimetric calculations for a theoretical $^{153}$Sm-HMFG1 radioimmunoconjugate

<table>
<thead>
<tr>
<th>$^{153}$Sm-HMFG1 uptake (% i.d./kg)</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>3.8</td>
<td>2.5</td>
<td>1.45</td>
</tr>
<tr>
<td>Mucosa</td>
<td>2.1</td>
<td>1.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Muscle</td>
<td>0.6</td>
<td>0.4</td>
<td>0.2</td>
</tr>
<tr>
<td>Cartilage</td>
<td>0.6</td>
<td>0.3</td>
<td>0.2</td>
</tr>
</tbody>
</table>

---

### Table 11: Dosimetric calculations for a theoretical $^{67}$Cu-HMFG1 radioimmunoconjugate

<table>
<thead>
<tr>
<th>$^{67}$Cu-HMFG1 uptake (% i.d./kg)</th>
<th>1 day</th>
<th>2 days</th>
<th>3 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tumor</td>
<td>4.2</td>
<td>3.0</td>
<td>1.9</td>
</tr>
<tr>
<td>Mucosa</td>
<td>2.3</td>
<td>1.9</td>
<td>0.8</td>
</tr>
<tr>
<td>Muscle</td>
<td>0.7</td>
<td>0.5</td>
<td>0.3</td>
</tr>
<tr>
<td>Cartilage</td>
<td>0.6</td>
<td>0.3</td>
<td>0.2</td>
</tr>
</tbody>
</table>

---

a) i.d., injected dose; b) Absorbed fraction; c) Corrected.
ACKNOWLEDGMENTS

REFERENCES


Radiolabeled Antibody Combined with External Radiotherapy for the Treatment of Head and Neck Cancer: Reconstruction of a Theoretical Phantom of the Larynx for Radiation Dose Calculation to Local Tissues

Anthony Maraveyas, Melvyn Myers, Nick Stafford, et al.


Access the most recent version of this article at: http://cancerres.aacrjournals.org/content/55/5/1020

Sign up to receive free email-alerts related to this article or journal.

To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.