

TOXICITY AND DOSIMETRY OF ^{177}Lu -DOTA-Y3-OCTREOTATE IN A RAT MODEL

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Radiolabeled somatostatin analogs have demonstrated effectiveness for targeted radiotherapy of somatostatin receptor-positive tumors in both tumor-bearing rodent models and humans. A radionuclide of interest for cancer therapy is reactor-produced ^{177}Lu ($t_{1/2} = 6.64$ d; β^- [100%]). The high therapeutic efficacy of the somatostatin analog ^{177}Lu -DOTA-Tyr³-octreotate (DOTA-Y3-TATE, where DOTA is 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid) was previously demonstrated in a tumor-bearing rat model (Erion et al., J. Nucl. Med. 1999;40:223P; de Jong et al., Int. J. Cancer, 2001; 92:628–633). In the current study, the toxicity and dosimetry of ^{177}Lu -DOTA-Y3-TATE were determined in both normal and tumor-bearing rats. Doses of ^{177}Lu -DOTA-Y3-TATE ranging from 0 to 123 mCi/kg were administered to rats and complete blood counts (CBCs) and blood chemistries were analyzed out to 6 weeks. No overt signs of toxicity were observed with ^{177}Lu -DOTA-Y3-TATE (i.e., lethargy, weight loss, scruffy coat or diarrhea) at any of the dose levels. Blood chemistries and CBCs were normal except for the white blood cell counts, which showed a dose-dependent decrease. The maximum tolerated dose was not reached at 123 mCi/kg. The biodistribution of ^{177}Lu -DOTA-Y3-TATE was determined in CA20948 rat pancreatic tumor-bearing rats, and the data were used to estimate human absorbed doses to normal tissues. The dose-limiting organ was determined to be the pancreas, followed by the adrenal glands. The absorbed dose to the rat CA20948 tumor was estimated to be 336 rad/mCi (91 mGy/MBq). These data demonstrate that ^{177}Lu -DOTA-Y3-TATE is an effective targeted radiotherapy agent at levels that show minimal toxicity in this rat model.

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Key words: targeted radiotherapy; lutetium-177; somatostatin; neuroendocrine cancer

The use of radiolabeled ligands that bind to somatostatin receptors (SSTRs) as targeted radiotherapy agents in humans is increasing.^{1–5} Preliminary results of clinical trials with ^{111}In -DTPA-OC, a diagnostic radiopharmaceutical approved by the United States Food and Drug Administration, have demonstrated evidence of tumor response to treatment.^{1,6} Reported responses include stable disease, but no significant tumor regressions or remissions. ^{90}Y -DOTA-Tyr³-octreotide (^{90}Y -DOTA-Y3-OC or ^{90}Y -DOTATOC or SMT487) is currently in clinical trials, with favorable results demonstrating significant tumor regressions.^{2–4} Preliminary data with ^{177}Lu -DOTA-tyrosine³-octreotate (^{177}Lu -DOTA-Y3-TATE) are presented in a recently published abstract and suggest a range of responses from tumor shrinkage (8/26) to stable disease (14/26) to partial remission (1/26) to tumor progression (3/26).⁵

The progress with somatostatin receptor ligands is very exciting; however, some questions remain to be answered. One issue is whether ^{90}Y is the best radionuclide for targeted radiotherapy of all tumor types and sizes.⁷ Yttrium-90 has a mean β^- -energy of 0.9 MeV with a maximum energy of 2.27 MeV and a maximum particle range of about 11 mm in tissue, making it an appropriate radionuclide for larger tumor burdens. A potential problem with ^{90}Y is that the high-energy β^- may also be the cause of the observed renal toxicity in humans treated with high doses of ^{90}Y -DOTATOC. This raises the question of whether or not other radionuclides with lower β^- energies may have utility for smaller tumors or micrometastatic disease.

An alternative radionuclide for targeted radiotherapy is ^{177}Lu ($t_{1/2} = 6.65$ d),⁸ which decays by lower energy β^- emissions (0.498 MeV [78.6%], 0.176 MeV [12.2%] and 0.385 [9.1%]).⁹ Lutetium-177 is produced at the University of Missouri Research Reactor (MURR) by the $^{176}\text{Lu}(n,\gamma)^{177}\text{Lu}$ reaction.¹⁰ In patients with ovarian cancer, the ^{177}Lu -labeled monoclonal antibody (Mab) CC49 showed considerable efficacy at controlling micrometastatic disease.¹¹ The groups at the University of Rotterdam and Mallinckrodt, Inc. have evaluated ^{177}Lu -labeled somatostatin analogs in a tumor-bearing rat model and demonstrated impressive therapeutic efficacy.^{12,13} Here we present data on the toxicity of ^{177}Lu -DOTA-tyrosine³-octreotate (^{177}Lu -DOTA-Y3-TATE; Fig. 1) in normal Lewis rats, as well as the estimated human absorbed doses of ^{177}Lu -DOTA-Y3-TATE to normal tissues and the dose to a rat tumor.

MATERIAL AND METHODS

General

All chemicals, unless otherwise stated, were purchased from Aldrich (Milwaukee, WI). Lutetium-177 was obtained from the University of Missouri Research Reactor in a specific activity of 20 Ci/mg.¹⁰ All solutions were prepared using ultrapure water (18 M Ω cm resistivity). Radio-thin layer chromatography (radio-TLC) detection was accomplished using a BIOSCAN (Washington, DC) System 200 Imaging Scanner. Radioactive samples were counted on a Beckman (Fullerton, CA) 8000- γ -counter.

Preparation of ^{177}Lu -DOTA-Y3-TATE

DOTA-Y3-TATE was synthesized as previously described.¹⁴ DOTA-Y3-TATE was labeled with $^{177}\text{LuCl}_3$ (0.05 M HCl) in 30 mM NaOAc/25 mM sodium ascorbate, pH 5.0, for 25 min at 80°C. Radiochemical purity was determined by radio-TLC with C18 plates developed in 70:30 MeOH:10% NH_4OAc (^{177}Lu -DOTA-Y3-TATE, $R_f = 0.8$; ^{177}Lu -acetate, $R_f = 0$). Radiochemical purity of ^{177}Lu -DOTA-Y3-TATE in all studies was >98%.

Abbreviations: DOTA, 1,4,7,10-tetraazacyclododecane-*N,N',N'',N'''*-tetraacetic acid; DTPA, diethylenetriaminepentaacetic acid; OC, octreotide; SSTR, somatostatin receptors; TATE, octreotate; TETA, 1,4,8,11-tetraazacyclotetradecane-*N,N',N'',N'''*-tetraacetic acid; Y3, tyrosine-3.

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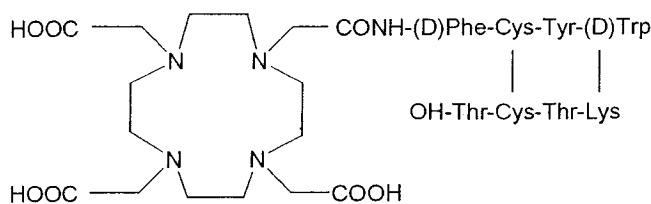


FIGURE 1 – Structure of DOTA-Y3-TATE.

Animal studies

All animal experiments were conducted in compliance with the Guidelines for the Care and Use of Research Animals established by Washington University's Animal Studies Committee. The rat pancreatic tumor CA20948¹⁵ was obtained from the Tumor Bank at Biomeasure (Hopkinton, MA). Adult male Lewis rats (190–240 g) were purchased from Charles River (Wilmington, MA). The CA20948 tumor was maintained by serial passage in animals. In rat experiments, male Lewis rats were injected with a 1 mm³ section of CA20948 tumor 10 days prior to treatment as previously described.¹⁶

Toxicity study

Doses of ¹⁷⁷Lu-DOTA-Y3-TATE (specific activity 0.89 mCi/ μ g) ranging from 0 to 130 mCi/kg in 5 toxicity groups (0–4.4, 16.3–18.2, 36.7–40.5, 64.7–71.2 and 102.2–122.6 mCi/kg; $n = 6$ in each group) were administered i.v. via the tail vein to normal rats. Toxicity was monitored by weight loss and gross physical appearance, as well as changes in hematologic, liver and kidney function compared with control rats ($n = 6$). In each group, anesthetized rats were weighed, and blood was removed by cardiac puncture before and during the experimental period. Toxicity analyses were performed by the Diagnostic Services Laboratory in the Department of Comparative Medicine at Washington University School of Medicine. The hematology analysis included white blood cell counts (WBCs), red blood cell counts (RBCs), platelet counts and measurement of hemoglobin, hematocrit and differential WBCs. Liver and kidney analysis included alanine aminotransferase (ALT), blood urea nitrogen (BUN) and creatinine. Gross necropsy examination of the respiratory, digestive, musculoskeletal, urinary and genital systems as well as the brain, thymus, spleen, lymph nodes, adrenals, pituitary and eye was performed on 2 animals from the highest dose group at the experimental endpoint of 38 days post injection. Histopathologic examination of the liver, kidney, pancreas and adrenal gland was also performed.

Rat dosimetry

The absorbed doses of ¹⁷⁷Lu-DOTA-Y3-TATE were obtained for normal tissues and a rat tumor using biodistribution data in CA20948 tumor-bearing rats, according to previously described methods.^{16–19} ¹⁷⁷Lu-DOTA-Y3-TATE (40 μ Ci [1.3 MBq]; 0.67 μ g) was injected i.v., and the rats bearing 0.5–3.5 g tumors were euthanized by Metofane overdose and cervical dislocation at 1, 3, 6, 14, 24, 48, 72, 96 and 168 hr (each group, $n = 5$) post injection. The rats in the 72 hr group were housed in metabolism cages to determine the percent injected dose (% ID) excreted in urine and feces at 1, 3, 18, 24, 48 and 72 hr.

Twenty-three organs and tissues (blood, lung, liver, spleen, kidney, bladder, muscle, fat, heart, brain, pituitary, bone, marrow [from femur], testes, prostate, adrenals, pancreas, thymus, tumor, stomach, small intestine, upper large intestine and lower large intestine) were harvested, and time-activity curves were generated for each of them. Additionally, the % ID/g and % ID/organ for each tissue were calculated. (Tables of % ID/g and % ID/organ with standard deviations for 23 tissues and 8 time points for ¹⁷⁷Lu-DOTA-Y3-TATE are available.) Uptake in the intestinal tract was

assumed to be due to fecal matter and not to receptor-specific uptake of the radiolabeled peptide. The cumulative activity (μ Ci/hr or kBq/hr) for each organ was determined by analytically integrating a mathematical function fitted by the least-square method on the data. This function was chosen to be a combination of exponentials.

Human absorbed dose estimates were calculated using measured residence times and the MIRD S-value for ¹⁷⁷Lu calculated with values supplied as supplementary information by the author of MIRDOSE 3.0.²⁰ The absorbed dose to the bladder was calculated assuming no voiding, and the dose to the kidney was calculated assuming homogenous distribution of the activity throughout the organ, a potential limitation of the MIRDOSE model. Tumor dosimetry was calculated taking into account all radiation emissions from ¹⁷⁷Lu, and the S-value for the tumor was calculated for a spherical tumor of unit density filled uniformly with activity. This approach assumed that tumors of similar size in different animals had similar uptake characteristics, and the resulting absorbed dose was the average of the doses absorbed by the individual tumors.

After reactor production, the ¹⁷⁷Lu contains 2 impurities, a small amount of ^{176m}Lu ($t_{1/2} = 3.64$ hr) and 0.02% of ^{177m}Lu ($t_{1/2} = 160.4$ days). Due to the short half-life of ^{176m}Lu and the delivery time for the isotope, its contribution to the absorbed dose estimates was not considered. The 0.02% presence of ^{177m}Lu, a value recently confirmed and published by the NIST,⁸ was considered, but the dose contribution of this contaminant was not included in our dosimetry calculations. It was assumed that this small impurity would only alter the absorbed dose estimates to the second decimal place and was easily within the reported standard deviations of the estimated absorbed doses.

Statistical analysis

To determine statistical significance in the biodistribution studies, a Student's *t*-test was performed with $p < 0.05$ being considered significantly different.

RESULTS

Toxicity study

The average weight of the treated rats increased similarly to that of the control animals, and they maintained a healthy physical appearance (with no sign of scruffy coat or diarrhea) over the experimental period. Throughout the examination period there were no significant changes in platelet counts, hemoglobin, hematocrit and differential WBCs (Table I). Levels of BUN, creatinine and ALT remained at baseline values, showing no overt liver and kidney toxicities. The hematology analysis values for the highest dose group (102.2–122.6 mCi/kg) are presented in Table I. WBC changes followed a dose-response pattern after administration (Fig. 2). In the lowest dose group (0–4.4 mCi/kg) there were no significant changes in WBCs over the experimental period (day –4, $11.95 \pm 4.51 \times 10^3/\text{mm}^3$; day 2, $14.0 \pm 4.38 \times 10^3/\text{mm}^3$; day 6, $12.8 \pm 2.64 \times 10^3/\text{mm}^3$; day 15, $11.8 \pm 1.64 \times 10^3/\text{mm}^3$; day 22, $11.4 \pm 0.60 \times 10^3/\text{mm}^3$; $p > 0.1$). The 3 intermediate groups exhibited a transient depression in WBCs at 6 days (16.3–18.2 mCi/kg, $11.3 \pm 2.43 \times 10^3/\text{mm}^3$; 36.7–40.5 mCi/kg, $8.9 \pm 1.15 \times 10^3/\text{mm}^3$; 64.7–71.2 mCi/kg, $5.83 \pm 1.34 \times 10^3/\text{mm}^3$; $p < 0.001$), but all recovered to baseline (day –4, $11.95 \pm 4.51 \times 10^3/\text{mm}^3$) levels by day 22. The 102.2–122.6 mCi/kg dose group showed the greatest neutropenic response; however, the WBCs recovered to baseline levels by day 35 (day –4, $14.15 \pm 2.98 \times 10^3/\text{mm}^3$; day 2, $8.4 \pm 4.41 \times 10^3/\text{mm}^3$; day 6, $3.9 \pm 1.8 \times 10^3/\text{mm}^3$; day 15, $5.6 \pm 2.47 \times 10^3/\text{mm}^3$; day 22, $8.2 \pm 2.00 \times 10^3/\text{mm}^3$; day 28, $10.5 \pm 1.99 \times 10^3/\text{mm}^3$; day 35, $11.4 \pm 1.58 \times 10^3/\text{mm}^3$; $p < 0.001$). In the gross necropsy and histopathologic examinations of selected tissues, there were no apparent lesions, showing the lack of gross or histologic signs of toxicity.

TABLE I – CHANGES IN HEMATOLOGIC, LIVER AND KIDNEY FUNCTION IN RATS TREATED WITH 102–122.6 mCi/kg ($n = 6$) OF ^{177}Lu -DOTA-Y3-TATE COMPARED WITH CONTROL RATS¹

Days after injection	RBC $10^6/\text{mm}^3$	Platelets $10^3/\text{mm}^3$	ALT U/L	Creatinine mg/dL	BUN mg/dL
	7.9 ± 0.46	774.8 ± 154.9	45.0 ± 25.0	0.5 ± 0.2	9.0 ± 2.1
2	8.1 ± 0.95	886.7 ± 492.8	60.3 ± 9.0	0.7 ± 0.1	10.7 ± 2.3
6	8.8 ± 0.76	1050.2 ± 392.76	58.2 ± 6.8	0.6 ± 0.1	14.2 ± 2.2
15	8.7 ± 0.88	654.8 ± 122.4	51.8 ± 9.8	0.5 ± 0.1	16.4 ± 3.5
22	8.1 ± 0.40	757.3 ± 108.0	44.5 ± 11.7	0.4 ± 0.0	15.1 ± 1.5
28	8.4 ± 0.33	909.2 ± 130.3	54.7 ± 17.3	0.5 ± 0.1	13.3 ± 1.5
35	9.3 ± 0.24	840.3 ± 142.9	68.0 ± 11.8	0.5 ± 0.2	17.3 ± 0.5

¹RBC, red blood cells; ALT, alanine aminotransferase; BUN, blood urea nitrogen.

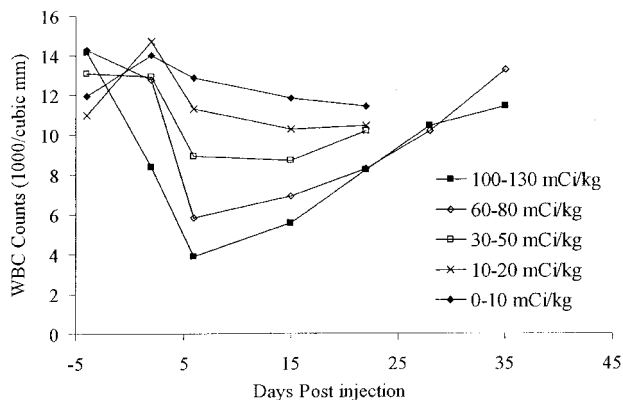


FIGURE 2 – White blood cell (WBC) counts ($10^3/\text{mm}^3$) in rats after administration of ^{177}Lu -DOTA-Y3-TATE in doses ranging from 0 to 123 mCi/kg. Error bars not shown for clarity.

Dosimetry

Human absorbed doses to normal organs from ^{177}Lu -DOTA-Y3-TATE were estimated from biodistribution data in CA20948 tumor-bearing rats. Biodistributions were performed at 1, 3, 6, 14, 24, 48, 72, 96 and 168 hr post injection, with the harvesting of 23 organs and tissues, a summary of which is shown in Table II. The rats for the 72 hr time point were housed in metabolism cages to determine %ID excreted in urine and feces at 1, 3, 18, 24, 48 and 72 hr (Table III). Total activity excreted in the urine by 72 hr was $68.74 \pm 2.37\%$, and the total fecal excretion was $15.84 \pm 3.11\%$. The human absorbed dose estimates to normal organs are presented in Table IV. The primary and secondary critical organs are the pancreas (11.12 ± 2.07 rad/mCi; 3.01 ± 0.56 mGy/MBq) and the adrenals (5.67 ± 0.86 rad/mCi; 1.53 ± 0.23 mGy/MBq), both somatostatin receptor-rich tissues. Primary excretion via the renal system resulted in an absorbed dose to the kidneys of 2.48 ± 0.17 rad/mCi (0.67 ± 0.05 mGy/MBq). In the CA20948 rat tumor, the average absorbed dose to the tumor was calculated to be 336 rad/mCi (91 mGy/MBq) for a single injection of ^{177}Lu -DOTA-Y3-TATE.

DISCUSSION

There have been several reports in recent years on targeted radiotherapy studies in tumor-bearing rodent models with radiolabeled somatostatin analogs labeled with ^{90}Y ^{21,22} and with alternative radionuclides such as ^{64}Cu ,^{16,19} ^{111}In ,^{1,6,23,24} ^{153}Sm ,²⁵ ^{161}Tb ,²⁶ ^{188}Re ²⁷ and ^{177}Lu .^{12,13} It has been demonstrated that with somatostatin-based peptides, changing the C-terminus from an alcohol (OC) to a carboxylic acid (TATE) increases uptake of these peptides in receptor-rich tissues.^{18,23,28} This can result in a higher dose to the tumor, while not dramatically affecting the absorbed doses to normal tissue.¹⁸ As a consequence, the most recent developments in the use of radiolabeled somatostatin analogs have focused on the use of the TATE derivative.^{12,13,19,25}

The radiotherapy data obtained in rats with ^{177}Lu -labeled somatostatin analogs has been particularly impressive.^{12,13} With ^{177}Lu -DOTA-Y3-TATE, the administration of 3×5 mCi over a 7-day period to tumor-bearing rats bearing 14-day-old CA20948 tumors resulted in a >95% decrease in tumor volume, with 66% of the animals remaining tumor-free 200 days after treatment.¹³ In the same study, a single 5 mCi administration of ^{177}Lu -DOTA-Y3-TATE in rats bearing 14-day-old CA20948 tumors resulted in tumor regression to <5% of the original tumor volume by day 14. In this group of rats >50% of the animals showed no palpable tumor mass and exhibited long-term survival of over 5 months. A multiple dose regimen of ^{177}Lu -DOTA-Y3-TATE (3×5 mCi at 30-day intervals) resulted in similar regression responses but with a greater percentage of animals surviving long term. De Jong *et al.*¹² observed a 100% cure rate in rats bearing small CA20948 tumors (<1 cm^3) after administering either 2 doses of 7.5 mCi ^{177}Lu -DOTA-Y3-TATE or a single dose of 15 mCi. In the same study using larger tumors (>1 cm^3), 40 and 50% cure rates were achieved in the groups that received 1 or 2 7.5 mCi injections of ^{177}Lu -DOTA-Y3-TATE, respectively. These data prompted the current study examining the maximum tolerated dose (MTD) levels and human absorbed dose estimates for ^{177}Lu -DOTA-Y3-TATE.

In the largest dose group examined in our study (102.2–122.6 mCi/kg), no abnormalities were found with regard to animal behavior (movement, sleeping, eating, posture) over the first 48 hr after injection. Moreover, the treated rats gained weight throughout the experiment and at no time had any overt physical signs of toxicity, such as lethargy, scruffy coat, >10% weight loss or diarrhea. Transient decreases in WBC levels were noted, but by day 35 after the first treatment the WBC counts returned to baseline values. Although not fully comprehensive, these toxicity data are encouraging since the MTD was not achieved, and it is apparent that larger quantities of radioactivity can probably be administered safely. It is particularly notable that in the same CA20948 tumor model a 100% cure rate was found with ^{177}Lu -DOTA-Y3-TATE by administering either 2 doses of 7.5 mCi or a single dose of 15 mCi.¹² The doses used in the toxicity study (102.2–122.6 mCi/kg) correspond to an administration of 22.9–24.4 mCi per animal, which are over 50–60% higher than doses administered in the successful therapy studies by de Jong *et al.* In addition, previous studies have demonstrated minimal toxicity of the unlabeled peptide DOTA-Y3-TATE at levels far in excess of those that would be used in humans.¹²

Previous studies have demonstrated the usefulness of using rat biodistribution in predicting human absorbed dose estimates. A preliminary evaluation of the absorbed doses in normal organs with ^{64}Cu -TETA-OC performed by averaging positron emission tomography (PET) data from 5 patients demonstrated that the urinary bladder was the dose-limiting organ, which was predicted from human absorbed dose measurements estimated from rat biodistribution and baboon PET imaging data.^{16,29} In the current study, the human absorbed estimates extrapolated from the biodistribution of ^{177}Lu -DOTA-Y3-TATE in rats showed that the primary and secondary critical organs were the pancreas (11.12 ± 2.07 rad/mCi; 3.01 ± 0.56 mGy/MBq) and

TABLE II – BIODISTRIBUTION AT 1, 3, 6, 24, 72 AND 168 HOURS OF ^{177}Lu -DOTA-Y3-TATE¹ IN SELECTED ORGANS

Organ	1 hr	3 hr	6 hr	24 hr	72 hr	168 hr
Blood	2.03 ± 0.42	0.31 ± 0.10	0.05 ± 0.05	0.03 ± 0.01	0.03 ± 0.00	0.01 ± 0.00
Lung	0.14 ± 0.01	0.08 ± 0.01	0.05 ± 0.00	0.04 ± 0.00	0.03 ± 0.01	0.02 ± 0.00
Liver	0.55 ± 0.23	0.34 ± 0.06	0.30 ± 0.03	0.28 ± 0.04	0.26 ± 0.08	0.18 ± 0.03
Spleen	0.02 ± 0.00	0.02 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.01 ± 0.01	0.01 ± 0.00
Kidney	1.92 ± 0.11	1.93 ± 0.11	1.63 ± 0.06	1.71 ± 0.13	1.55 ± 0.09	0.94 ± 0.05
Muscle	1.25 ± 0.25	0.27 ± 0.03	0.08 ± 0.05	0.12 ± 0.01	0.09 ± 0.02	0.07 ± 0.02
Heart	0.04 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
Pituitary	0.03 ± 0.01	0.05 ± 0.01	0.04 ± 0.00	0.04 ± 0.01	0.03 ± 0.00	0.03 ± 0.01
Bone	11.96 ± 0.96	9.28 ± 1.34	5.47 ± 3.08	6.70 ± 0.75	5.03 ± 0.52	4.20 ± 0.19
Adrenal	0.31 ± 0.06	0.31 ± 0.09	0.30 ± 0.02	0.21 ± 0.03	0.14 ± 0.02	0.11 ± 0.02
Small intestine	0.89 ± 0.44	0.93 ± 0.48	0.73 ± 0.19	0.33 ± 0.06	0.19 ± 0.04	0.15 ± 0.05
Upper large intestine	0.77 ± 0.14	1.32 ± 0.47	2.70 ± 0.76	0.47 ± 0.12	0.16 ± 0.03	0.13 ± 0.01
Lower large intestine	2.87 ± 0.46	2.82 ± 0.27	4.89 ± 1.94	3.04 ± 0.65	0.87 ± 0.16	0.68 ± 0.21
Pancreas	10.49 ± 1.91	9.20 ± 1.42	6.43 ± 2.76	2.25 ± 0.32	1.80 ± 0.36	1.07 ± 0.10
Tumor	3.92 ± 3.84	5.92 ± 3.00	5.97 ± 4.04	6.13 ± 2.84	2.86 ± 1.77	0.65 ± 0.35

Data for 15 of 23 organs are given as % injected dose (ID) per organ ± SD ($n = 5$).¹The %ID/g data are available as supplementary information. Data for 14, 48 and 96 hr are not shown.

TABLE III – EXCRETION OF ^{177}Lu -DOTA-Y3-TATE IN CA20948 TUMOR-BEARING LEWIS RATS ($N = 5$) AT SELECTED TIME POINTS

	% Injected dose ± SD				
	1 hr	1–3 hr	3–24 hr	24–48 hr	1–72 hr total
Urine	20.91 ± 18.56	15.46 ± 19.15	26.13 ± 17.95	2.54 ± 0.64	68.74 ± 2.37
Feces	0.004 ± 0.012	0.004	6.12 ± 1.79	3.17 ± 0.94	15.84 ± 3.11

TABLE IV – HUMAN ABSORBED DOSE ESTIMATES OF ^{177}Lu -DOTA-Y3-TATE OBTAINED IN CA20948-BEARING RATS

Organ	Rad/mCi ± SD	mGy/MBq ± SD
Adrenals	5.670 ± 0.86	1.533 ± 0.233
Lower large intestine	3.289 ± 1.17	0.889 ± 0.316
Small intestine	0.172 ± 0.06	0.047 ± 0.015
Stomach	0.461 ± 0.09	0.125 ± 0.023
Upper large intestine	0.440 ± 0.10	0.119 ± 0.028
Heart wall	0.042 ± 0.01	0.011 ± 0.002
Kidneys	2.477 ± 0.17	0.670 ± 0.047
Liver	0.086 ± 0.02	0.023 ± 0.005
Lungs	0.029 ± 0.01	0.008 ± 0.002
Muscle	0.038 ± 0.01	0.010 ± 0.004
Pancreas	11.12 ± 2.07	3.006 ± 0.561
Red marrow	0.360 ± 0.05	0.097 ± 0.014
Bone surfaces	1.926 ± 0.23	0.521 ± 0.061
Spleen	0.074 ± 0.02	0.020 ± 0.004
Bladder wall	1.322 ± 0.16	0.357 ± 0.042
Total body	0.116 ± 0.03	0.031 ± 0.007

the adrenals (5.67 ± 0.86 rad/mCi; 1.53 ± 0.23 mGy/MBq), both somatostatin receptor-rich tissues. It was further demonstrated that the primary excretion route was via the renal system, which resulted in an absorbed dose to the kidneys of 2.48 ± 0.17 rad/mCi (0.67 ± 0.05 mGy/MBq). It should be noted that the kidney uptake of ^{177}Lu -DOTA-Y3-TATE can be significantly reduced by 40% by the coinjection of 400 mg/kg D-lysine.¹² The transient depression in WBCs may also be indicative of the dose delivered to the bone marrow causing toxicity. A multiple dose regimen might significantly reduce the absorbed doses to nontarget organs by allowing the delivery of a consistent amount of tolerable radiation over an extended period to the tumor, while allowing intermittent recovery of nontarget tissues. It is also important to note that hepatobiliary and renal clearance of many radiopharmaceuticals,³⁰ as well as receptor concentrations and subtype expression,³¹ vary widely from rodents to humans and that primate and actual human doses may be improved over dose estimates from animal models. With this in mind, in humans the primary and secondary tissue may not be the pancreas and the adrenals.

In the de Jong *et al.*¹² study, a single 15 mCi dose of ^{177}Lu -DOTA-Y3-TATE resulted in a 100% cure in 200–300 g

rats bearing small tumors, which would suggest a dose of over 2 Ci of ^{177}Lu -DOTA-Y3-TATE for clinical therapy trials in humans. In the ongoing clinical trials the patients are scheduled to receive a maximum cumulative dose of only 600–800 mCi.⁵ In these human trials, 26 patients with progressive disease were given 300 mCi of ^{177}Lu -DOTA-Y3-TATE with amino acid infusion to reduce the kidney dose.⁵ Of the 26 patients, 31% experienced nausea, 9% vomiting and 11% mild abdominal discomfort. Additionally, of the 26 patients, 5 presented with mild leukocytopenia, 3 with mild thrombocytopenia and 10 with mild anemia. At the time of presentation none of the patients had received their maximum cumulative dose of 600–800 mCi and had shown a range of responses from tumor shrinkage (8/26) to stable disease (14/26) to partial remission (1/26) to tumor progression (3/26).

The absorbed dose to the CA20948 tumor from ^{177}Lu -DOTA-Y3-TATE calculated from the rat biodistribution was 336.46 ± 203 rad/mCi (90.94 ± 54.9 mGy/MBq); a similar value of 96 mGy/MBq was reported by de Jong *et al.*¹² This value is higher than that calculated for ^{64}Cu -DOTA-Y3-TATE (33.2 rad/mCi) but is considerably lower than that calculated for ^{90}Y -DOTA-Y3-TATE (1753 rad/mCi).¹⁸ Comparing the estimated absorbed doses for ^{177}Lu -DOTA-Y3-TATE with ^{64}Cu -DOTA-Y3-TATE, the dose imparted to the kidney was ~5 times higher (2.48 ± 0.17 vs. 0.48 ± 0.16 rad/mCi) but imparted ~10-fold more dose (336 rad/mCi vs. 33.24 rad/mCi) to the CA20948 tumor. When comparing the absorbed dose to the kidney between ^{177}Lu -DOTA-Y3-TATE and ^{90}Y -DOTA-Y3-TATE, the dose imparted to the kidney was ~3 times lower (2.48 ± 0.17 vs. 7.29 ± 1.22 rad/mCi), with a ~5-fold reduction in dose (336 rad/mCi vs. 1753 rad/mCi) to the CA20948 tumor. These data suggest that although ^{177}Lu -DOTA-Y3-TATE appears to have distinct advantages over ^{64}Cu -DOTA-Y3-TATE, ^{177}Lu -DOTA-Y3-TATE and ^{90}Y -DOTA-Y3-TATE may be more comparable with respect to their tumor and normal tissue toxicity.

Doses of ^{177}Lu -DOTA-Y3-TATE ranging from 0 to 123 mCi/kg were administered to normal rats and complete blood counts (CBCs) and blood chemistries were analyzed out to 6 weeks. No overt signs of toxicity were observed with ^{177}Lu -DOTA-Y3-TATE at any of the dose levels. We further estimated human absorbed radiation doses to normal tissues and the absorbed dose to the rat CA20948 tumor for ^{177}Lu -DOTA-Y3-TATE, for which

the adrenals and pancreas were determined to be the dose-limiting organs. Since previous studies have demonstrated that ¹⁷⁷Lu-DOTA-Y3-TATE is effective in causing tumor regression of CA20948 tumors in rats, these toxicity and dosimetry data suggest that ¹⁷⁷Lu-DOTA-Y3-TATE may be an effective targeted radiotherapy agent at levels that show minimal toxicity in humans. These data, in conjunction with the human trial data,⁵ further suggest that this radiopharmaceutical may be at least as effective as the ⁹⁰Y-labeled somatostatin analogs currently in use.

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