

# Radioimmunotherapy with $^{111}\text{In}/^{90}\text{Y}$ -2IT-BAD-m170 for Metastatic Prostate Cancer<sup>1</sup>

Robert T. O'Donnell,<sup>2</sup> Sally J. DeNardo, Aina Yuan, Sui Shen, Carol M. Richman, Primo N. Lara, Irwin J. Griffith, Desiree S. Goldstein, David L. Kukis, Gerard S. Martinez, Gary R. Mirick, Gerald L. DeNardo, and Frederick J. Meyers

Department of Internal Medicine, Division of Hematology and Oncology, University of California Davis Medical Center, Sacramento, California 95816, and Biomira, Inc., Edmonton, Alberta, Canada T6N 1H1 [I. J. G.]

## ABSTRACT

**Purpose:** Over 31,000 Americans die of androgen-independent metastatic prostate cancer each year. New strategies that do not involve hormonal manipulation but instead recognize the biochemical and molecular characteristics of prostate cancer are needed. Radioimmunotherapy (RIT) uses a tumor-specific monoclonal antibody to deliver systemic, targeted radiation to cancer. The objectives of this Phase I study of  $^{111}\text{In}$ -2IT-BAD-m170 (for imaging) and  $^{90}\text{Y}$ -2IT-BAD-m170 (for therapy) were to determine the toxicity and maximum tolerated dose (MTD), the specificity for targeting metastatic prostate cancer, and the efficacy for palliation of pain.

**Experimental Design:** M170 is a mouse monoclonal antibody that targets adenocarcinomas. Patients with adequate renal and liver function, rising prostate-specific antigen, and androgen-independent metastatic prostate cancer were eligible. After estimation of dosimetry and pharmacokinetics with  $^{111}\text{In}$ -2IT-BAD-m170, a single dose of  $^{90}\text{Y}$ -2IT-BAD-m170 (0.185, 0.370, 0.555, or 0.740 GBq/m<sup>2</sup>) was administered to cohorts of three patients. Pain was assessed objectively by questionnaires before and for 8 weeks after RIT; weekly prostate-specific antigen levels were obtained for 2 months after RIT.

**Results:** The MTD of  $^{90}\text{Y}$ -2IT-BAD-m170 was 0.740 GBq/m<sup>2</sup> for patients that had up to 10% of the axial skeleton

involved with prostate cancer. Toxicity was almost exclusively confined to reversible myelosuppression. Metastatic prostate cancer was targeted by  $^{111}\text{In}$ -2IT-BAD-m170 in all 17 patients. The mean radiation dose delivered to 39 bone and 18 nodal metastases by  $^{90}\text{Y}$ -2IT-BAD-m170 was 10.5 Gy/GBq (range 2.8–25.1). Thirteen of 17 patients reported pain before  $^{90}\text{Y}$ -2IT-BAD-m170; 7 of these 13 had a partial or complete resolution of pain that lasted an average of 4.3 weeks.

**Conclusions:** This study determined the MTD of  $^{111}\text{In}/^{90}\text{Y}$ -2IT-BAD-m170 in patients with metastatic prostate cancer. The drugs were well tolerated, targeted metastases, and temporarily palliated pain.

## INTRODUCTION

Metastatic prostate cancer causes the deaths of over 31,000 Americans each year (1). Debilitation and pain exact a great toll on quality of life. Androgen ablation is the only treatment that prolongs life after metastasis has occurred, and even then, the 2.5-year survival is only 50% (2). The accumulation of molecular alterations in androgen-independent prostate cancer may explain its resistance to systemic chemotherapy (3). New, innovative therapies for metastatic prostate cancer are needed. RIT<sup>3</sup> is systemic radiation therapy in which a MAb labeled with high-energy, short-range radioisotopes specifically concentrates in disseminated tumor, thereby minimizing concomitant radiation of normal tissues. Because prostate cancer usually metastasizes to bone and lymph nodes, is relatively responsive to radiation, and occurs in a patient population that has had little if any exposure to cytotoxic chemotherapy, it is a disease in which RIT should be tested.

Although impressive results have been reported using RIT for advanced hematological malignancies (4), success in solid tumors has been more limited. Radiolabeled MAbs have been used to detect metastatic prostate cancer, but demonstrating therapeutic impact on the disease itself has been elusive. Foci of metastatic prostate cancer were detected by  $^{111}\text{In}$ -DTPA-PSA 399, a murine IgG, in 9 of 10 patients despite the potential for binding to circulating PSA (5).  $^{111}\text{In}$ -CYT-356 (ProstaScint), a murine IgG1 MAb conjugated with the linker-chelator 64K-DTPA, has been used to target a  $M_r$  100,000 glycoprotein (6). In a therapeutic trial, 58% of patients had detectable uptake of  $^{111}\text{In}$ -CYT-356 in at least one site, however no complete or partial responses were achieved by treatment with a single dose

Received 10/15/00; revised 2/13/01; accepted 2/14/01.

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.

<sup>1</sup> Supported by grants from the Cancer Research Institute and NCI PO1-CA47829. The authors acknowledge the support of the Veteran's Administration Northern California Healthcare System (to R. T. O.) and of the American Cancer Society Clinical Research Training Grant (to P. N. L.).

<sup>2</sup> To whom requests for reprints should be addressed, at University of California, Davis, 1508 Alhambra Boulevard, Sacramento, CA 95816. Phone: (916) 734-3787; Fax: (916) 451-2857; E-mail: rtodonnell@ucdavis.edu.

<sup>3</sup> The abbreviations used are RIT, radioimmunotherapy; MAb, monoclonal antibody; PSA, prostate-specific antigen; MTD, maximum tolerated dose; HAMA, human antimouse antibody; CT, computed tomography; BAD, 2-[p-(bromoacetamido)benzyl]-1,4,7,10-tetraazacyclododecane-*N,N',N''*-tetraacetic acid; 2IT, 2-iminothiolane; DTPA, diethylenetriaminepentaacetic acid; DOTA, 1,4,7,10-tetra-azacyclododecane *N,N',N'',N'''*-tetraacetic acid.

Table 1 Patient characteristics

Cohort group	Age	Pre-RIT PSA <sup>a</sup>	Prior chemotherapy	Prior radiotherapy <sup>b</sup>	% of axial skeleton positive for PC on bone scan
1A	58	246	N	L	0
1A	69	37	N	L	4.2
1A	76	29	N	D	4.7
2A	69	174	N	L	8.4
2A	56	155	N	D	6.5
2A	64	40	N	N	2.0
3A	45	61	N	N	0
3A	64	67	N	N	9.7
3A	60	99	N	L	8.3
3A	79	157	N	L	2.0
3A	68	128	N	N	2.3
1B	60	68	N	D	23.8
1B	70	194	N	L	21.0
1B	63	94	Y	D	12.3
2B	67	1033	N	L + D	15.6
2B	62	732	Y	D	18.2
2B	57	223	Y	L	15.4

<sup>a</sup> Normal range for PSA, 0–4.

<sup>b</sup> L, local radiation therapy to the prostate bed; D, distant radiation therapy to metastatic prostate cancer; N, no prior radiation therapy.

of <sup>90</sup>Y-CYT-356 (7). CC49, a murine IgG1, binds a high molecular weight mucin antigen found on many adenocarcinomas. <sup>131</sup>I-CC49 relieved pain in 6 of 10 patients with prostate cancer, but none met the study criteria for a PSA or radiographic response, and all developed an immune response against the antibody (8). KC4 is a murine IgG3 MAb that reacts with a high molecular weight adenocarcinoma-associated membrane and cytoplasmic antigen; <sup>90</sup>Y-KC4 was administered to seven patients with prostate cancer, and mild to moderate pain relief was achieved in some patients (9).

m170 (170H.82) is an IgG1 MAb produced by immunization of mice with a synthetic asialo GM1 terminal disaccharide similar to the cancer-associated Thomsen-Friedenreich disaccharide (10), and m170 can bind to a variety of human adenocarcinoma specimens (11). In this study, <sup>111</sup>In-2IT-BAD-m170 and <sup>90</sup>Y-2IT-BAD-m170 were administered to 17 patients to evaluate the safety, type, and degree of toxicity; the MTD; the specificity for the targeting of androgen-independent metastatic prostate cancer; and the potential of RIT for palliation of pain.

## MATERIALS AND METHODS

**Patient Characteristics.** All 17 patients had progressive prostate cancer that was androgen-independent, Table 1. Androgen independence was defined as a PSA >20 ng/ml and rising at least 3 months after orchiectomy or chemical hormonal ablation. The average age of patients was 64 years. Three of 17 patients had received chemotherapy before RIT. Eight patients had received radiation therapy to the prostatic bed and six patients had received external beam radiotherapy to sites of metastatic disease. Patients were eligible if they had absolute neutrophil and platelet counts of at least 1,500 and 120,000/mm<sup>3</sup>, respectively; bilateral bone marrow biopsies with <25% prostate cancer involvement; HAMA titer <5 µg/ml; aspartate aminotransferase <3 times normal; creatinine clearance at least 60 ml/min; Karnofsky performance score at least 70%; no anticancer therapy for at least 4 weeks before <sup>111</sup>In-2IT-BAD-m170; no evidence of a second neoplasm; measurable disease

by X-ray, CT, MRI, or bone scan; and <25% of the axial or total skeleton positive for prostate cancer on a bone scan obtained within 1 month of RIT. Before therapy, patients signed an informed consent approved by the University of California, Davis, Human Subjects and Radiation Use Committees under an Investigational New Drug authorization from the Food and Drug Administration.

**Preparation of <sup>111</sup>In/<sup>90</sup>Y-2IT-BAD-m170.** m170 (Bio-mira, Edmonton, Canada) was >95% monomeric IgG by PAGE and met United States Food and Drug Administration guidelines. To prepare <sup>111</sup>In/<sup>90</sup>Y-2IT-BAD-m170, m170 was first conjugated to the bifunctional chelating agent BAD via 2IT (Sigma Chemical Co., St. Louis, MO) to prepare 2IT-BAD-m170 (12). <sup>90</sup>Y (Pacific Northwest National Laboratory, Richland, WA) and 2IT-BAD-m170 were combined in 0.4 M ammonium acetate (pH 7) for 15–30 min at 37°C (13). DTPA (Sigma Chemical Co.) was added (final concentration, 0.1 M) to scavenge nonspecifically bound <sup>90</sup>Y, then <sup>90</sup>Y-2IT-BAD-m170 was purified by Sephadex G25 molecular sieving chromatography and formulated in 4% human serum albumin/saline at ~0.037 GBq/ml. <sup>111</sup>In-2IT-BAD-m170 was prepared in a manner similar to <sup>90</sup>Y-2IT-BAD-m170, except that <sup>111</sup>In (Nordion, Kanata, Ontario, Canada) was combined with 2IT-BAD-m170 in 0.1 M ammonium acetate (pH 5), and EDTA (Fisher Scientific, Fair Lawn, NJ) was used to scavenge nonspecifically bound <sup>111</sup>In.

<sup>111</sup>In/<sup>90</sup>Y-2IT-BAD-m170 was examined for structural and functional integrity by cellulose acetate electrophoresis (Gelman Sciences, Inc., Ann Arbor, MI) and molecular sieving high-performance liquid chromatography (Beckman, Fullerton, CA), as described previously (14). The immunoreactivity of <sup>111</sup>In-2IT-BAD-m170 or <sup>90</sup>Y-2IT-BAD-m170 were determined by an assay in which they competed with <sup>125</sup>I-m170 for binding to rabbit-anti-m170 idiotype antibody. Dilutions of decayed <sup>111</sup>In-2IT-BAD-m170 or <sup>90</sup>Y-2IT-BAD-m170 and <sup>125</sup>I-m170 (4 ng) were incubated for 2 h in a tube coated with rabbit-anti-m170 idiotype antibody. The amount of <sup>125</sup>I-m170 bound in the ab-

Table 2 Cohort groups<sup>a</sup>

Cohort group A: ≤10% of axial skeleton involved by prostate cancer on bone scan		Cohort group B: >10% but ≤25% of axial skeleton involved by prostate cancer on bone scan	
	GBq/m <sup>2</sup>		
na	0.185	1B	
1A	0.370	2B	
2A	0.555	3B	
3A	0.740	na	

<sup>a</sup> Patients were assigned to groups “A” or “B” by the percentage of bone involved by prostate cancer.

sence of <sup>111</sup>In-2IT-BAD-m170 (or <sup>90</sup>Y-2IT-BAD-m170) denoted maximal binding. The amount of decayed <sup>111</sup>In-2IT-BAD-m170 or <sup>90</sup>In-2IT-BAD-m170 required to reduce <sup>125</sup>I-m170 binding to one-half the maximal binding (ED<sub>50</sub>) was determined (a smaller ED<sub>50</sub> indicated higher immunoreactivity). A parallel study was performed in which the ED<sub>50</sub> of an m170 reference standard was assessed. The percent immunoreactivity of <sup>111</sup>In-2IT-BAD-m170 and <sup>90</sup>In-2IT-BAD-m170 were calculated by relating their immunoreactivity to that of the reference standard. An ionization chamber dose calibrator (Capintec CRC-12; Capintec, Inc., Pittsburgh, PA) was used to measure <sup>111</sup>In/<sup>90</sup>In-2IT-BAD-m170 doses. <sup>90</sup>In-2IT-BAD-m170 had 97.5 ± 1.6% radiochemical purity on monomeric m170 as assessed by cellulose acetate electrophoresis and corroborated by high-performance liquid chromatography. The immunoreactivity of <sup>90</sup>In-2IT-BAD-m170 was 65 ± 19%; a mean specific activity of 0.11 ± 0.021 GBq <sup>90</sup>Y/mg m170 was achieved. <sup>111</sup>In-2IT-BAD-m170 had 100 ± 0.01% radiochemical purity on monomeric m170. The immunoreactivity of <sup>111</sup>In-2IT-BAD-m170 was 70 ± 20%; the mean specific activity was 0.072 ± 0.022 GBq <sup>111</sup>In/mg m170.

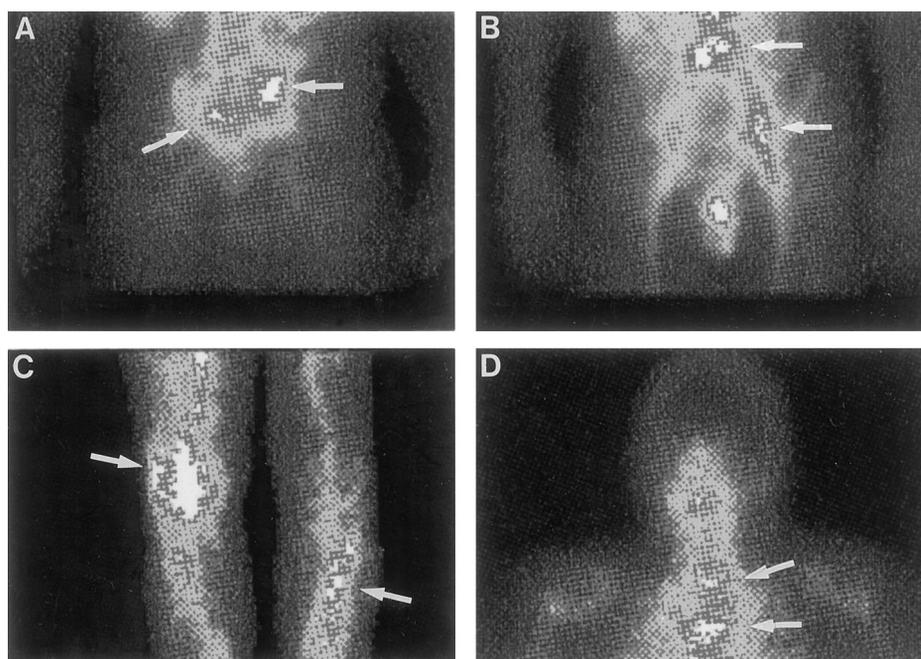
**Study Design.** The study objectives were to determine the toxicity and MTD of <sup>90</sup>In-2IT-BAD-m170 and to detect specific targeting of metastatic prostate cancer with <sup>111</sup>In-2IT-BAD-m170. Palliation of disease-related symptoms and changes in the serum tumor marker, PSA, were also measured. Patients were assigned to dose escalation cohort groups (Table 2). As measured by bone scan, patients with ≤10% bone involved by prostate cancer received a higher dose (Cohort A) than those with >10% up to 25% involvement (Cohort B). Radiopharmaceuticals were administered 30 min after premedication with acetaminophen (650 mg), diphenhydramine (50 mg), and dexamethasone (20 mg), given to preclude the possibility (albeit unlikely) of an allergic reaction to the murine antibody. Five mg of unmodified m170 was given to reduce nonspecific adherence of m170. Diphenhydramine and acetaminophen were also given 3 and 6 h after radiopharmaceutical administration. RIT was administered in an outpatient clinic. Unconjugated m170 and radiopharmaceuticals were infused at ~0.5 mg/min. Vital signs were monitored at least every 15 min before, during, and for 2 h after m170 infusion and then every 30 min for 2 additional hours.

<sup>90</sup>Y has potent β but no γ emissions, therefore, <sup>111</sup>In-2IT-BAD-m170 was used as a surrogate for <sup>90</sup>In-2IT-BAD-m170. Each patient received <sup>111</sup>In-2IT-BAD-m170 (0.185 GBq/m<sup>2</sup>) for

imaging to calculate pharmacokinetics and dosimetry for <sup>90</sup>In-2IT-BAD-m170, which was given 1 week later (15). Imaging was done immediately and 4 h after the <sup>111</sup>In-2IT-BAD-m170 infusion, and on 3 additional days during the subsequent week. Blood and urine samples were obtained and the radioactivity measured. Normal tissue dose limits were set, so that blood-to-marrow, and liver, lung, and kidney radiation doses would not exceed 1.35 Gy/GBq, 15.0 Gy, 12.5 Gy, and 10.0 Gy, respectively. If imaging and dosimetry were less than the normal organ radiation dosimetry limits, and uptake was seen in prostate cancer metastases, the treatment dose of <sup>90</sup>Y-2IT-BAD-m170 (0.185, 0.370, 0.555, or 0.740 GBq/m<sup>2</sup>) was administered after premedication, as described above. None of 17 patients in the study had radiation dosimetry that exceeded the bone marrow or organ dosimetry limits that would have required a dose reduction.

Liver and kidney function tests, CBC, HAMA, and PSA levels were obtained before <sup>111</sup>In-2IT-BAD-m170. Weekly PSA levels were taken for at least 8 weeks after RIT. Weekly CBCs were done after RIT for at least 8 weeks and until blood counts had recovered to baseline or grade 1 toxicity. Renal and liver function tests and a HAMA assay were repeated 4–6 weeks after RIT. The MTD was determined by grade 3 or 4 thrombocytopenia for >14 or 7 days, respectively, or grade 3 granulocytopenia that lasted for >10 days, or Grade 4 nonhematological toxicity. History and physical examination was performed before each radiopharmaceutical infusion, at 1 week after RIT, and monthly for 3 months after the final dose. Three independent, objective quantitative assessments of pain and quality of life were obtained: a monthly “Fact-L” quality of life questionnaire; a daily “Pain Medication Diary”; and a weekly “Brief Pain Inventory.” The 1988 National Cancer Institute Common Toxicity Criteria were used to classify data. Serum HAMA was measured by an enzyme-linked immunoabsorbent assay using microtiter plates coated with m170 as described previously (16). In this Phase I study, response was assessed by pain control and by PSA values, which were required to decrease by at least 50% for at least 1 month to qualify as a partial response. PSA velocities were calculated by linear regression using PSA values obtained after the prostate cancer had become androgen-independent up until the day of <sup>111</sup>In-2IT-BAD-m170 administration and were obtained for the month after <sup>90</sup>Y-2IT-BAD-m170 for post-RIT PSA velocity.

**Pharmacokinetics and Radiation Dosimetry.** Blood samples were obtained for analysis of radiopharmaceutical content immediately, at 4 h, and on 3 additional days during the next week. Blood radioactivity was counted in a gamma well counter and compared with a standard prepared from the batch of radiopharmaceutical that was injected into the patient. The percentage of the injected dose in the blood was calculated for each sample using the body weight to estimate blood volume. Pharmacokinetic data were obtained as described previously; a biexponential function was used to determine cumulated activity in blood (17). Planar images of opposing views were acquired immediately, at 4 h, and 3 times during the week after administration of <sup>111</sup>In-2IT-BAD-m170 (18, 19). Cumulated activity was obtained for liver, lung, kidney, and whole body using a monoexponential analysis and converted to radiation dose using the Medical Internal Radiation Dose formula, Medical Internal



*Fig. 1* Gamma camera imaging of <sup>111</sup>In-2IT-BAD-m170 uptake into sites of metastatic prostate cancer in four patients (arrows). *A*, day-3 posterior pelvic view of massive retroperitoneal adenopathy that was also seen on CT and palpable by physical examination. *B*, day-3 anterior pelvic view of retroperitoneal and left inguinal adenopathy that was also seen on CT. *C*, day-2 anterior view of the lower femur, knee, and tibia showing uptake of <sup>111</sup>In-2IT-BAD-m170 into sites of prostate cancer seen on bone scan and X-ray as extensive, mottled, osteoblastic lesions. *D*, day-6 anterior chest view showing uptake of <sup>111</sup>In-2IT-BAD-m170 into sternal and manubrial metastasis that were also seen on bone scan.

Radiation Dose *S* values, and reference-man masses. For tumor dosimetry to be accurate, tumors must have had a measured volume of at least 10 ml. Tumor diameters were determined using calipers or radiographic measurements; volume was determined as described previously (20). Metastases of sufficient volume must also have been seen on the <sup>111</sup>In-2IT-BAD-m170 study in order for their radiation dosimetry to be calculated. Fifty-seven sites of metastatic prostate cancer met the criteria for dosimetric analysis. The bone marrow radiation dose was determined as the marrow-to-marrow, nonpenetrating radiation dose extrapolated from the uptake of <sup>111</sup>In-2IT-BAD-m170 as imaged in three lumbar vertebrae (21, 22). The radiation dose from blood to marrow was also calculated, as described previously:  $D_{\text{total}} = 0.25 \bar{A}_{\text{blood}} \Delta_{\text{np}}$ , where  $\bar{A}_{\text{blood}}$  is the cumulated activity in 1 ml of blood and  $\Delta_{\text{np}}$  is the mean energy emitted per nuclear transition for nonpenetrating <sup>90</sup>Y emissions. The constant 0.25 was used to account for the difference between the specific activities of marrow and of circulating blood (17, 22, 23).

## RESULTS

**Dose.** Quantitative imaging of <sup>111</sup>In-2IT-BAD-m170 was used to predict dosimetry of its therapeutic counterpart, <sup>90</sup>Y-2IT-BAD-m170. The radiation doses determined by <sup>111</sup>In-2IT-BAD-m170 dosimetry for all 17 patients were far below the radiation dose limits set forth (dose limits for bone marrow, liver, lungs, and kidney were 1.35 Gy/GBq, 15 Gy, 12.5 Gy, and 10 Gy, respectively). Therefore, all doses of <sup>90</sup>Y-2IT-BAD-m170 were given on the GBq/m<sup>2</sup> basis as prescribed for their cohort group (Table 2). Doses of 0.185 GBq/m<sup>2</sup> were escalated to 0.740 GBq/m<sup>2</sup> in three patient cohort groups, except for cohort 3A, where an additional two patients were treated to more firmly ascertain the MTD.

**Prostate Cancer Targeting.** <sup>111</sup>In-2IT-BAD-m170 was infused, and during the subsequent week, patients were imaged

*Table 3* <sup>90</sup>Y-2IT-BAD-m170 dosimetry in 17 patients with androgen-independent metastatic prostate cancer

	Gy/GBq (mean ± SD)	Range (Gy/GBq)
Tumor (bone), <i>n</i> = 39	10.86 ± 5.00	2.80–21.10
Tumor (nodal), <i>n</i> = 18	10.05 ± 5.64	3.20–25.10
Liver	3.95 ± 0.67	2.62–4.98
Lung	1.75 ± 0.38	1.26–2.49
Kidney	2.04 ± 0.58	1.29–3.64
Body	0.62 ± 0.02	0.58–0.64
Blood-to-bone marrow	0.69 ± 0.13	0.49–0.97
Bone marrow by imaging of 3 lumbar vertebrae	1.67 ± 0.34	1.10–2.40

with a gamma camera and single-photon emission CT. Bone and/or soft tissue prostate cancer metastasis were successfully targeted by <sup>111</sup>In-2IT-BAD-m170 in all 17 patients, Fig. 1.

**Pharmacokinetics and Radiation Dosimetry.** The radiation dose targeted to bone and lymph node metastases by the radioactive MAb was much higher than doses delivered to normal organs (Table 3). The mean radiation dose to 57 evaluable prostate cancer metastases by <sup>90</sup>Y-2IT-BAD-m170 was 10.5 ± 5.0 Gy/GBq. The mean radiation dose to both bone and nodal prostate cancer metastasis was greater than 10 Gy/GBq, and some sites of metastatic disease received doses exceeding 20 Gy/GBq. Dosimetry revealed a good therapeutic ratio for tumor:normal organs, with liver consistently being the highest normal organ, with a mean dose of 3.95 ± 0.67 Gy/GBq.

**Toxicity.** Myelosuppression occurred as a result of RIT in patients treated with <sup>90</sup>Y-2IT-BAD-m170; however, it was brief and reversible (Table 4). Myelotoxicity was modest after a single dose of <sup>90</sup>Y-2IT-BAD-m170 up to 0.740 GBq/m<sup>2</sup>. No patient experienced bleeding while thrombocytopenic or re-

Table 4 Duration of myelotoxicity and bone marrow dosimetry

Cohort Group <sup>a</sup> (GBq/m <sup>2</sup> )	Gy/GBq→bone marrow	cGy→bone marrow	Granulocytopenia k/mm <sup>3</sup> (days)			Thrombocytopenia k/mm <sup>3</sup> (days)		
			<1500	<1000	<500	<75	<50	<25
1A (0.370)	1.8	129	0	0	0	0	0	0
1A (0.370)	1.4	121	8	0	0	6	0	0
1A (0.370)	1.7	125	34	20	7	14	14	0
2A (0.555)	1.8	190	27	20	8	30	21	7
2A (0.555)	1.8	223	7	7	0	7	0	0
2A (0.555)	1.6	199	16	0	0	7	0	0
3A (0.740)	2.2	289	11	7	0	14	10	0
3A (0.740)	2.0	274	7	3	0	10	10	5
3A (0.740)	1.2	214	0	0	0	16	0	0
3A (0.740)	2.4	327	23	2	0	21	11	1
3A (0.740)	1.9	295	7	7	0	7	3	0
1B (0.185)	1.3	59	0	0	0	0	0	0
1B (0.185)	1.4	59	0	0	0	0	0	0
1B (0.185)	1.6	63	0	0	0	0	0	0
2B (0.370)	1.8	142	0	0	0	0	0	0
2B (0.370)	1.1	71	0	0	0	0	0	0
2B (0.370)	1.6	128	15	2	0	0	0	0

<sup>a</sup> A cohorts, ≤10% of axial skeleton involved by prostate cancer on bone scan; B cohorts, >10% but ≤25% axial skeleton involved by prostate cancer on bone scan.

quired a platelet transfusion. No patient had significant infection, neutropenic fever, or nonhematological toxicity, Table 4. Nonhematological toxicity included a grade 1 upper respiratory tract infection (1 patient), gastrointestinal toxicity (grade 2 diarrhea after imaging and grade 2 diarrhea after therapy; 1 patient), and grade 2 constipation with pain (likely related to narcotics; 1 patient). Some patients reported mild fatigue that was less than grade 1. There was no hepatotoxicity or elevation of liver function tests in any patient. HAMA became positive (>5 µg/ml) in 12 of 17 patients but had no adverse effect. HAMA was not an unexpected occurrence after mouse MAb-based RIT in patients with adenocarcinoma.

**Response.** Although this was a Phase I study designed to determine the toxicity and MTD, PSA and pain control were analyzed. It is notable that the PSA velocity observed before RIT was interrupted by a brief period of stable or decreasing PSA velocity for the month after a single dose of <sup>90</sup>Y-2IT-BAD-m170 in 7 of 17 (41%) patients (Table 5).

The pain scores from the Pain Medication Diary and Fact-L questionnaire (obtained before RIT and weekly after RIT), objectively confirmed the substantial decrease in pain after <sup>90</sup>Y-2IT-BAD-m170 that was reported by the patients in clinic. Of 17 patients, 13 reported pain immediately before RIT. Complete resolution of pain after RIT occurred in 5 of 13 (38%) patients; an additional 2 of 13 (15%) patients had at least a 50% reduction in pain. The responding patients had at least a 50% reduction of pain for a mean of 4.3 weeks. Of the seven patients that had improved pain control, two had some pain relief after the imaging dose with additional improvement after <sup>90</sup>Y-2IT-BAD-m170; the other five responding patients had improved pain control only after RIT with <sup>90</sup>Y-2IT-BAD-m170.

## DISCUSSION

Patients with androgen-independent metastatic prostate cancer have limited therapeutic options. Standard therapy frequently provides incomplete palliation of symptoms. RIT has

the potential to target therapeutic radiation specifically to metastatic tumors in bone and soft tissues. This Phase I study showed that metastatic prostate cancer could be specifically targeted, and that some patients had pain relief and/or a decrease in PSA velocity, with modest, manageable toxicity. Therefore, this study provides the basis for subsequent Phase I RIT studies that can evaluate methods to enhance further the antitumor response.

m170 reacts with many types of adenocarcinoma, including >90% of breast cancers, however, it had not been used previously for RIT of prostate cancer (10, 11). <sup>90</sup>Y is a pure β emitter with a high therapeutic impact and prolonged tumor retention time. Most investigators use β-emitting radionuclides, such as <sup>131</sup>I, <sup>90</sup>Y, and <sup>67</sup>Cu to deliver more uniform radiation to the tumor and to alleviate the problem of poor tumor penetration by the MAb (24, 25). BAD contains the macrocyclic chelator DOTA, which stably binds radiometals (26). The resulting drugs, <sup>111</sup>In-2IT-BAD-m170 and <sup>90</sup>Y-2IT-BAD-m170, were used in this Phase I clinical trial to deliver DOTA-chelated <sup>111</sup>In and <sup>90</sup>Y to androgen-independent metastatic prostate cancer.

<sup>111</sup>In-2IT-BAD-m170 targeted metastatic prostate cancer in all 17 patients. Normal tissue clearance and tumor uptake of radiolabeled-MAb differ among patients; quantitative imaging provides a noninvasive method for obtaining radiation dosimetric information for the radiopharmaceuticals. Radionuclide images depict the distribution of radiolabeled antibodies *in vivo*. In this study, <sup>111</sup>In-2IT-BAD-m170 was used to predict the dosimetry of its therapeutic counterpart, <sup>90</sup>Y-2IT-BAD-m170. The treatment radionuclide, <sup>90</sup>Y, cannot be imaged by standard means because it has no γ emissions; so <sup>111</sup>In was used as a surrogate to predict the dosimetry of identically chelated <sup>90</sup>Y. For additional safety, the injected radioactivity (GBq) was not allowed to be more than that which would cause a greater radiation dose (Gy) to normal organs than the limits defined in the protocol. The mean radiation dose to tumor was 2.7 times higher than the mean radiation dose to the highest normal organ

Table 5 Pre-RIT and post-RIT PSA velocity and pain-control response in 17 patients with androgen-independent metastatic prostate cancer

Cohort <sup>a</sup>	<sup>90</sup> Y dose (GBq/m <sup>2</sup> )	Pre-RIT PSA (mo) <sup>b</sup>	Pre-RIT PSA velocity <sup>c</sup>	Post-RIT PSA velocity <sup>c</sup>	Pain-control response <sup>d</sup>	Pain-control response CR + PR <sup>d</sup> (wk)
1A	0.370	3	2.04	0.53	NR	0
1A	0.370	7	0.17	-1.43	CR	1 + 4
1A	0.370	2	0.20	-0.04	CR	2 + 0
2A	0.555	11	0.58	-2.07	PR	0 + 6
2A	0.555	6	0.63	1.69	NA <sup>e</sup>	NA
2A	0.555	5	0.12	0.21	NA <sup>e</sup>	NA
3A	0.740	11	0.16	0.58	NA <sup>e</sup>	NA
3A	0.740	11	0.20	-0.90	CR	5 + 0
3A	0.740	8	0.43	2.07	CR	5 + 0
3A	0.740	5	0.63	1.64	NR	NA
3A	0.740	7	0.51	-0.18	NR	NA
1B	0.185	8	0.25	0.58	PR	0 + 2
1B	0.185	6	1.03	3.90	NR	0
1B	0.185	4	0.48	0.66	CR	4 + 1
2B	0.370	8	3.17	14.00	NA <sup>e</sup>	NA
2B	0.370	6	3.05	32.20	NR	0
2B	0.370	2	2.40	0.38	NR	0

<sup>a</sup> Bone involvement for cohort A, ≤10%; bone involvement for cohort B, >10% ≤25%.

<sup>b</sup> Number of months of PSA values from which pre-RIT PSA velocity was calculated.

<sup>c</sup> Velocity = slope of PSA versus time. Post-RIT PSA velocity is for 1 month after RIT.

<sup>d</sup> NR, no response; CR, complete response; PR, partial response; NA, not applicable.

<sup>e</sup> Did not complain of pain prior to RIT.

(liver), demonstrating specific targeting of prostate cancer. Pharmacokinetics and dosimetry in the prostate cancer patients imaged with <sup>111</sup>In-2IT-BAD-m170 were similar to those of breast cancer patients imaged with <sup>111</sup>In-2IT-BAD-m170. In patients with breast cancer, dosimetry calculated by <sup>111</sup>In-2IT-BAD-m170 pharmacokinetics for liver, lung, kidney, and tumors were 4.8, 1.7, 1.9, and 19.2 Gy/GBq, respectively. In each study, only small mg doses of m170 were required to achieve good biodistribution of the radiopharmaceuticals. Preliminary results of an ongoing study using high-dose <sup>90</sup>Y-2IT-BAD-m170 with peripheral blood stem cell support in patients with metastatic breast cancer are encouraging (27).

A number of clinical studies have assessed the utility of RIT for prostate cancer. <sup>111</sup>In-Capromab pendetide (ProstaScint; Cytogen Corporation, Princeton NJ), uses the mouse IgG1 CYT-356 MAb to target prostate-specific membrane antigen (28). ProstaScint has demonstrated the ability to detect prostate cancer and assists in the staging of patients (29). ProstaScint is approved for use in newly diagnosed patients who are at high risk for occult metastatic disease and in patients with rising PSA after radical prostatectomy. Because of its targeting ability for prostate cancer, a therapeutic clinical trial with <sup>90</sup>Y-CYT-356 was performed (7). In 12 patients, 7 had at least one site of disease imaged by <sup>111</sup>In-CYT-356. The dose-limiting toxicity for <sup>90</sup>Y-CYT-356 was myelosuppression, and the MTD was 9 mCi/m<sup>2</sup> (0.33 GBq/m<sup>2</sup>). Slovin *et al.* (30) used an interesting approach in which patients were pretreated with IFN-γ to increase expression of the TAG-72 target antigen. Then 14 patients received <sup>131</sup>I-CC49 (75 mCi/m<sup>2</sup>; 2.78 GBq/m<sup>2</sup>). The dose-limiting toxicity was grade 3 or 4 thrombocytopenia, which occurred in the majority of patients. Six patients had a flare of baseline pain, and in 4 patients, pain decreased.

RIT has had increasingly encouraging results in a variety of other solid tumors. In a pilot clinical trial using the <sup>131</sup>I-chimeric

L6 MAb, objective responses occurred in 6 of 10 patients with metastatic breast cancer that had failed chemotherapy (31). Promising results in patients with breast cancer have also been reported using mouse and humanized BrE-3 MAbs that target an epitope on the peptide core of MUC-1 (15, 32). Lechner *et al.* (33) conducted a dosimetric study of <sup>111</sup>In/<sup>90</sup>Y-PA-DOTA-CC49 in 12 patients with gastrointestinal tumors. The mean tumor and liver doses calculated for <sup>90</sup>Y-CC49 were 7.2 and 10.1 Gy/GBq, respectively. Up to a mean of 30 Gy were delivered to tumors in this study. Juweid *et al.* (34) used high-dose <sup>131</sup>I-MN-14, an anticarcinoembryonic antigen F(ab)<sub>2</sub>, with peripheral blood stem cell support in a study of patients with metastatic medullary thyroid cancer, and antitumor responses were achieved. High therapeutic indices were achieved with mean tumor doses of 6.3 Gy/GBq, compared with liver, lung, and kidney doses of 0.6, 0.6, and 0.7 Gy/GBq, respectively.

Although <sup>90</sup>Y-2IT-BAD-m170 produced some myelotoxicity in patients that received less than the MTD, there was a more consistent degree of myelotoxicity as the dose of <sup>90</sup>Y increased to 0.740 GBq/m<sup>2</sup>. Additional dose escalation was not required to define the toxicity from the radiopharmaceutical. The toxicity detected was almost exclusively myelosuppression, and additional dose escalation may have resulted in even lower blood counts that would have been unnecessary to define the toxicity profile. Because the toxicity was confined to myelosuppression, substantially increased doses of <sup>90</sup>Y-2IT-BAD-m170 could be used if hematopoietic stem cell support was to be used. Bone involvement with prostate cancer has theoretical implications for myelotoxicity subsequent to bone marrow radiation; however, at up to 25% bone involvement, no association with increased toxicity could be ascribed. No obvious pretreatment parameter predicted hematological toxicity in this relatively small patient population. The dosimetry determined in this study

showed good tumor:normal organ dose ratios and this may explain the lack of significant nonhematological toxicity.

Alterations in PSA velocity rather than actual PSA values were used to describe changes. No patient had a reduction of PSA that would qualify as a partial response. However, all of the patients had increasing PSA levels at the time of study entry, and the PSA velocity transiently decreased in 7 of 17; PSA velocity actually had a negative slope after RIT in 5 of 17 patients. Ultimately, the PSA velocities and pain returned. There were pain-control responses at each  $^{90}\text{Y}$  dose level tested; however, there were too few patients in each group to firmly determine a dose-response relationship.

Targeted delivery of radiation to metastatic prostate cancer using antitumor MAbs is evolving from a research endeavor to a practical therapeutic modality. This Phase I study was done mainly to determine the MTD; single  $^{90}\text{Y}$  doses up to 0.740 GBq/m<sup>2</sup> could be given safely to patients that had <10% axial skeletal involvement by prostate cancer.  $^{111}\text{In}/^{90}\text{Y}$ -2IT-BAD-m170 can be safely administered with minimal toxicity confined to reversible myelosuppression. The liver received the highest (but still modest) normal-organ radiation dose, but no hepatotoxicity was manifested. HAMA can occur but had no adverse impact. This study showed that a single, modest dose of  $^{90}\text{Y}$ -2IT-BAD-m170 has the potential to decrease pain and PSA velocity. In the future, multiple dose regimens may use cyclosporin A to suppress HAMA formation. The combination of synergistic chemotherapy with RIT in future clinical trials may lead to more durable responses.

## REFERENCES

- Greenlee, R., Murray, T., Bolden, S., and Wingo, P.A. Cancer Statistics, 2000. *CA Cancer J. Clin.*, 50: 7–33, 2000.
- Denis, L., Churner De Mora, J. L., Bono, A., Sylvester, R., Whelan, P., Newline, D., and Depauw, M. Goserelin acetate and flutamide versus bilateral orchiectomy: a Phase III EORTC trial (30853). *Urology*, 42: 119–130, 1993.
- Lara, P. N., Kung, H. J., Gumerlock, P. H., and Meyers, F. J. Molecular biology of prostate carcinogenesis. *Crit. Rev. Oncol. Hematol.*, 32: 197–208, 1999.
- DeNardo, G. L., O'Donnell, R. T., Oldham, R. K., and DeNardo, S. J. A revolution in the treatment of non-Hodgkin's lymphoma. *Cancer Biother. Radiopharm.*, 13: 213–223, 1998.
- Meyers, F. J., DeNardo, S. J., Macey, D. J., White, R. D., and Unger, M. Development of monoclonal antibody imaging of metastatic prostatic carcinoma. *Prostate*, 14: 209–220, 1989.
- Bander, N. H. Current status of monoclonal antibodies for imaging and therapy of prostate cancer. *Semin. Oncol.*, 21: 607–612, 1994.
- Deb, N., Goris, M., Trisler, K., Fowler, S., Saal, J., Ning, S., Becker, M., Marquez, C., and Knox, S. Treatment of hormone-refractory prostate cancer with 90-Y-CYT-356 monoclonal antibody. *Clin. Cancer Res.*, 2: 1289–1297, 1996.
- Meredith, R. F., Bueschen, A. J., Khazaeli, M. B., Plott, W. E., Grizzle, W. E., Wheeler, R. H., Schlom, J., Russell, C. D., Liu, T., and LoBuglio, A. F. Treatment of metastatic prostate carcinoma with radio-labeled antibody CC49. *J. Nucl. Med.*, 35: 1017–1022, 1994.
- Abdel-Nabi, H., Spaulding, E. F., Derby, L., Lamonica, D., and Glenn, S. Treatment of refractory prostate carcinoma with 90Y KC4. *J. Nucl. Med.*, 36: 2–13, 1995.
- McEwan, A. J. B., MacLean, G. D., Hooper, H., Sykes, T., McQuarrie, S. A., Goldberg, L., Bodnar, D. M., Lloyd, S. L., and Noujaim, A. A. MAb 170H.82: an evaluation of a novel panadenocarcinoma monoclonal antibody labeled with  $^{99}\text{Tc}^m$  and with  $^{111}\text{In}$ . *Nucl. Med. Commun.*, 13: 11–19, 1992.
- Howell, L. P., DeNardo, S. J., Levy, N., Lund, J., and DeNardo, G. L. Immunohistochemical staining of metastatic ductal carcinomas of the breast by monoclonal antibodies used in imaging and therapy: a comparative study. *Int. J. Biol. Markers*, 10: 126–135, 1995.
- McCall, M. J., Diril, H., and Meares, C. F. Simplified method for conjugating macrocyclic bifunctional chelating agents to antibodies via 2-iminothiolane. *Bioconjug. Chem.*, 1: 222–226, 1990.
- Kukis, D. L., DeNardo, S. J., DeNardo, G. L., O'Donnell, R. T., and Meares, C. F. Optimized conditions for chelation of yttrium-90-DOTA immunoconjugates. *J. Nucl. Med.*, 39: 2105–2110, 1998.
- DeNardo, S. J., DeNardo, G. L., Kukis, D. L., Shen, S., Kroger, L. A., DeNardo, D. A., Goldstein, D. S., Mirick, G. R., Salako, Q., Mausner, L. F., Srivastava, S. C., and Meares, C. F.  $^{67}\text{Cu}$ -2IT-BAT-Lym-1 pharmacokinetics, radiation dosimetry, toxicity, and tumor regression in patients with lymphoma. *J. Nucl. Med.*, 40: 302–310, 1999.
- DeNardo, S. J., Kramer, E. L., O'Donnell, R. T., Richman, C. M., Salako, Q. A., Shen, S., Noz, M., Glenn, S. D., Ceriani, R. L., and DeNardo, G. L. Radioimmunotherapy for breast cancer using indium-111/yttrium-90 BrE-3: results of a Phase I clinical trial. *J. Nucl. Med.*, 38: 1180–1185, 1997.
- DeNardo, G. L., Mirick, G. R., Kroger, L. A., O'Donnell, R. T., Meares, C. F., and DeNardo, S. J. Antibody responses to macrocycles in lymphoma. *J. Nucl. Med.*, 37: 451–456, 1996.
- DeNardo, G. L., Mahe, M. A., DeNardo, S. J., Macey, D. J., Mirick, G. R., Erwin, W. D., and Groch, M. W. Body and blood clearance and marrow radiation dose of  $^{131}\text{I}$ -Lym-1 in patients with B-cell malignancies. *Nucl. Med. Commun.*, 14: 587–595, 1993.
- Erwin, W. D., Groch, M. W., Macey, D. J., DeNardo, G. L., DeNardo, S. J., and Shen, S. A radioimmunotherapy and MIRD dosimetry treatment planning program for radioimmunotherapy. *Nucl. Med. Biol.*, 23: 525–532, 1996.
- Shen, S., DeNardo, G. L., DeNardo, S. J., Salako, Q., Morris, G., Banks, D., Yuan, A., and DeNardo, D. A. Dosimetric evaluation of Copper-64 in Copper-67-2IT-BAT-Lym-1 for radioimmunotherapy. *J. Nucl. Med.*, 37: 146–150, 1996.
- DeNardo, G. L., O'Donnell, R. T., Shen, S., Kroger, L. A., Yuan, A., Meares, C. F., Kukis, D. L., and DeNardo, S. J. Radiation dosimetry for  $^{90}\text{Y}$ -2IT-BAD-Lym-1 extrapolated from pharmacokinetics using  $^{111}\text{In}$ -2IT-BAD-Lym-1 in patients with non-Hodgkin's lymphoma. *J. Nucl. Med.*, 41: 952–958, 2000.
- DeNardo, G. L., DeNardo, S. J., Kukis, D. L., O'Donnell, R. T., Shen, S., Goldstein, D. S., Kroger, L. A., Salako, Q., DeNardo, D. A., Mirick, G. R., Mausner, L. F., Srivastava, S. C., and Meares, C. F. Maximum tolerated dose of  $^{67}\text{Cu}$ -2IT-BAT-Lym-1 for fractionated radioimmunotherapy of non-Hodgkin's lymphoma: a pilot study. *Anticancer Res.*, 18: 2779–2788, 1998.
- Lim, S.-M., DeNardo, G. L., DeNardo, D. A., Shen, S., Yuan, A., O'Donnell, R. T., and DeNardo, S. J. Prediction of myelotoxicity using radiation doses to marrow from body, blood, and marrow sources. *J. Nucl. Med.*, 38: 1374–1378, 1997.
- Dillman, L. T. Radionuclide decay schemes and nuclear parameters for use in radiation-dose estimation, part 2. MIRD Pamphlet No. 6, pp. 5–32. New York: Society of Nuclear Medicine, 1970.
- Wessels, B. W., Vessella, R. L., Palme, D. F., Berkopec, J. M., Smith, G. K., and Bradley, E. W. Radiobiological comparison of external beam irradiation and radioimmunotherapy in renal cell carcinoma xenografts. *Int. J. Radiat. Oncol. Phys.*, 17: 1257–1263, 1989.
- Langmuir, V. K., McGann, J. K., Buchegger, F., and Sutherland, R. M. The effect of antigen concentration, antibody valency and size, and tumor architecture on antibody binding in multicell spheroids. *Nucl. Med. Biol.*, 18: 753–764, 1991.
- Deshpande, S. V., DeNardo, S. J., Kukis, D. L., Moi, M. K., McCall, M. J., DeNardo, G. L., and Meares, C. F. Yttrium-90-labeled monoclonal antibody for therapy: labeling by a new macrocyclic chelating agent. *J. Nucl. Med.*, 30: 473–479, 1989.
- Richman, C. M., DeNardo, S. J., O'Donnell, R. T., Goldstein, D. S., Shen, S., Kukis, D. L., Kroger, L. A., Yuan, A., Boniface, G. R., Griffith, I. J., and DeNardo, G. L. Dosimetry-based therapy in metastatic

- breast cancer patients using <sup>90</sup>Y monoclonal antibodies 170H.82 with autologous stem cell support and cyclosporin A. *Clin. Cancer Res.*, 5: 3243s–3248s, 1999.
28. Petronis, J., Regan, F., and Lin, K. Indium-111 Capromab Pentetide (ProstaScint) imaging to detect recurrent and metastatic prostate cancer. *Clin. Nucl. Med.*, 23: 672–677, 1998.
29. Manyak, M. Clinical applications of radioimmunoscinigraphy with prostate-specific antibodies for prostate cancer. *Cancer Control*, 5: 493–499, 1998.
30. Slovin, S., Scher, H., Divgi, C., Reuter, V., Sgouros, G., Moore, M. A., Weingard, K., Pettengall, R., Imbriaco, M., El-Shirbiny, A. M., Finn, R., Bronstein, J., Brett, C., Milenic, D. E., Dnistrian, A., Shapiro, L., Schlom, J., and Larson, S. M. Interferon- $\gamma$  and monoclonal antibody <sup>131</sup>I-labeled CC49; outcomes in patients with androgen-independent prostate cancer. *Clin. Cancer Res.*, 4: 643–651, 1998.
31. DeNardo, S. J., O'Grady, L. F., Richman, C. M., Goldstein, D. S., O'Donnell, R. T., DeNardo, D. A., Kroger, L. A., Lamborn, K. R., Hellstrom, K. E., Hellstrom, I., and DeNardo, G. L. Radioimmunotherapy for advanced breast cancer using I-131-ChL6 antibody. *Anti-cancer Res.*, 17: 1745–1752, 1997.
32. Cagnoni, P. J., Ceriani, R. L., Cole, W. C., Johnson, R., Quaife, R., Nieto, Y., Matthes, S. M., Schpall, E. J., Bearman, S. I., McDerm, H. S., Cook, B., Peterson, J., Blank, E., and Jones, R. B. High-dose radioimmunotherapy with <sup>90</sup>Y-hu-BrE-3 followed by autologous hematopoietic stem cell support (AHSCS) in patients with metastatic breast cancer. *Cancer Biother. Radiopharm.*, 14: 318, 1999.
33. Leichner, P. K., Akabani, G., Colcher, D., Harrison, K. A., Hawkins, W. G., Eckblade, M., Baranowska-Kortylewicz, J., Augustine, S. C., Wisecarver, J., and Tempero, M. A. Patient-specific dosimetry of indium-111- and yttrium-90-labeled monoclonal antibody CC49. *J. Nucl. Med.*, 38: 512–516, 1997.
34. Juweid, M. E., Hajjar, G., Stein, R., Sharkey, R. M., Herskovic, T., Swayne, L. C., Suleiman, S., Pereira, M., Rubin, A. D., and Goldenberg, D. M. Initial experience with high-dose radioimmunotherapy of metastatic medullary thyroid carcinoma using <sup>131</sup>I-MN-14 F(ab)<sub>2</sub> anti-carcinoembryonic antigen (CEA) monoclonal antibody and autologous hematopoietic stem rescue (AHSCR). *J. Nucl. Med.*, 41: 93–103, 2000.

# Clinical Cancer Research

## Radioimmunotherapy with $^{111}\text{In}/^{90}\text{Y}$ -2IT-BAD-m170 for Metastatic Prostate Cancer

Robert T. O'Donnell, Sally J. DeNardo, Aina Yuan, et al.

*Clin Cancer Res* 2001;7:1561-1568.

**Updated version** Access the most recent version of this article at:  
<http://clincancerres.aacrjournals.org/content/7/6/1561>

**Cited articles** This article cites 31 articles, 12 of which you can access for free at:  
<http://clincancerres.aacrjournals.org/content/7/6/1561.full#ref-list-1>

**Citing articles** This article has been cited by 11 HighWire-hosted articles. Access the articles at:  
<http://clincancerres.aacrjournals.org/content/7/6/1561.full#related-urls>

**E-mail alerts** [Sign up to receive free email-alerts](#) related to this article or journal.

**Reprints and Subscriptions** To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at [pubs@aacr.org](mailto:pubs@aacr.org).

**Permissions** To request permission to re-use all or part of this article, use this link  
<http://clincancerres.aacrjournals.org/content/7/6/1561>.  
Click on "Request Permissions" which will take you to the Copyright Clearance Center's (CCC) Rightslink site.