# Phase I Evaluation of J591 as a Vascular Targeting Agent in Progressive Solid Tumors

Michael J. Morris,[1,5] Neeta Pandit-Taskar,[2,3] Chaitanya R. Divgi,[2,3,5] Samantha Bender,[2] Joseph A. O'Donoghue,[4] Angelo Nacca,[3] Peter Smith-Jones,[2,3,5] Lawrence Schwartz,[3] Susan Slovin,[1,5] Ronald Finn,[3] Steven Larson,[2,3,5] and Howard I. Scher[1,5]

**Abstract**

**Purpose:** The antibody J591 targets the external domain of prostate-specific membrane antigen, which is expressed in the neovasculature of nonprostate solid tumors. This phase I trial tested the hypothesis that J591 could be used as a vascular targeting platform for patients with nonprostate solid tumors.

**Experimental Design:** Patients with progressive solid tumors were eligible. Twenty patients, divided into six dosage cohorts of 3 to 6 patients each, were treated every 3 weeks to a maximum of four doses using either 5, 10, 20, 40, 60, or 100 mg of J591 antibody. Two milligrams of antibody were labeled with 10 mCi of indium-111.

**Results:** Patients with a wide variety of solid tumors were tested; all had good tumor localization. No dose-limiting toxicities were observed. The serum clearance rate decreased with increasing antibody mass, likely a result of early hepatic uptake of antibody. Half-life for each successive cohort was 0.71, 0.84, 1.86, 1.83, 3.32, and 3.56 days. Hepatic saturation seemed to occur by 60 mg. Seventeen of 18 (94%) patients with soft tissue disease on standard scans showed uptake in the soft tissues on antibody scans as did 6 of 6 patients with bone disease.

**Conclusions:** The tumoral neovasculature of a variety of solid tumors can be selectively and safely targeted using J591. In planning for future studies using J591 as a radiation delivery platform, an antibody mass of 60 mg should be considered, as it would seem to minimize the radiation delivered to the liver while minimizing the radiation dose to bone.

Prostate-specific membrane antigen (PSMA) is a 100-kDa transmembrane glycoprotein found in a variety of normal human tissues, such as prostate epithelium, the central nervous system, and the proximal gastrointestinal tract (1–3). To date, PSMA has primarily been used to treat and image prostate cancer. For imaging, it is the basis of the ProstaScint scan, a Food and Drug Administration–approved study for detecting nodal metastases (4). For therapy, it is the target of unlabeled, radiolabeled, and chemoconjugated antibodies (5–7). PSMA is also expressed in the neovascular endothelium of most solid tumors, including lung, colon, breast, renal, transitional cell, and pancreas cancers. It is not expressed in normal vasculature (8, 9). Preclinical data suggest that PSMA may be a key regulator of angiogenic activity, and PSMA-null animals have severely impaired angiogenesis (10). As such, it is a promising means of selectively targeting neoplastic tissue in patients who have nonprostate solid tumors.

J591 (MLN591; Millennium Pharmaceuticals) is a monoclonal IgG1 molecule that targets the external domain of PSMA (11), modified to reduce its immunogenicity by replacing individual amino acid sequences in the antibody variable domains (DeImmunisation, Biovation Ltd.). Previous studies have shown that J591 localizes to metastatic prostate cancer, stimulates antibody-dependent cellular cytotoxicity, and induces PSA declines (5). Phase I studies have also shown that J591 conjugated to indium-111 ($^{111}$In), yttrium-90, and lutetium-177 is safe, targets prostate cancer metastases to bone and soft tissues, and, when labeled with therapeutic radiation doses, can induce biochemical and objective responses (6, 12).

In the current phase I trial, we sought to establish the feasibility of targeting the neovasculature of nonprostate solid tumors using a PSMA-directed monoclonal antibody and to define the relationship between antibody mass, biodistribution, pharmacokinetics, and toxicity.

## Materials and Methods

### Eligibility

Eligible patients had histologically documented, advanced nonprostate solid tumors. Patients were required to have disease

**Authors' Affiliations:** [1]Genitourinary Oncology Service, Department of Medicine, [2]Nuclear Medicine Service, Departments of [3]Radiology, and [4]Medical Physics, Memorial Sloan-Kettering Cancer Center; and [5]Department of Medicine, Weill Medical College of Cornell University, New York, New York

Received 12/12/06; revised 1/24/07; accepted 2/19/07.

**Grant support:** National Cancer Institute grants CA102544 and CA05826, Prostate Cancer Foundation, Sacerdote Fund, PepsiCo Foundation for Prostate Cancer, Mr. William H. Goodwin and Mrs. Alice Goodwin and the Commonwealth Cancer Foundation for Research, and The Experimental Therapeutics Center of Memorial Sloan-Kettering Cancer Center.

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.

**Note:** Presented in part at the annual meeting of the AACR, 2003.

**Requests for reprints:** Michael J. Morris, Genitourinary Oncology Service, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, Box 444, New York, NY 10021. Phone: 646-422-4469; Fax: 212-988-0701; E-mail: morrism@mskcc.org.



doi:10.1158/1078-0432.CCR-06-2935

progression documented by WHO criteria, Karnofsky performance status >60%, WBC count >3,500/μL, platelet count >150,000/μL, bilirubin <2.0 mg/dL, aspartate aminotransferase <3 × the upper limit of normal, creatinine <2.0 mg/dL or creatinine clearance >60 mL/min, and prothrombin time <14.5 s.

Patients could not have received anticancer therapy for at least 4 weeks before entry into the trial. All patients signed informed consent. The protocol was approved by the Institutional Review Board of Memorial Sloan-Kettering Cancer Center.

### Treatment

Twenty patients were enrolled and treated in cohorts of three to six. Cohorts were given 5, 10, 20, 40, 60, or 100 mg of antibody. Each administered dose of J591, infused at a rate of 5 mg/min, contained 2 mg of antibody radiolabeled with 10 mCi of $^{111}$In conjugated using the chelator 1,4,7,10-tetra-azacylododecane $N,N',N'',N'''$-tetraacetic acid (DOTA). Patients were treated every 21 days (which constituted the length of one cycle) for a maximum of four treatments.

A history, physical exam, toxicity check, complete blood count, and chemistries were done on the day of treatment, day 8, and any day between days 15 and 19.

### Labeling procedure

Two milligrams of antibody in each dose were labeled with 10 mCi of $^{111}$In by adding the radionuclide (in diluted HCl) to the ammonium acetate buffered DOTA-J591 (supplied by BZL Biologics, Inc.). The volume of $^{111}InCl_3$ was buffered by adding 1 mol/L ammonium acetate (pH 7.0) and allowed to react at 37°C for 20 min with 225 μL DOTA-hu-J591. The reaction mixture was separated on a 20 mL polyacrylamide P6 gel column (Bio-Rad) equilibrated with 6 × 5 mL of sterile 1% human serum albumin in PBS. The $^{111}$In-DOTA-hu-J591 fraction was eluted with 3 mL of 1% human serum albumin in PBS. The purified $^{111}$In-DOTA-hu-J591 was terminally filtered to achieve sterility.

### End points

***Toxicity.*** National Cancer Institute Common Toxicity Criteria version 2 was used. Infusion-related rigors and chills were expected treatment-related side effects and were treated with meperidine, diphenhydramine, and acetaminophen if they occurred. If any grade 3 or 4 drug-related toxicities occurred, the patient was taken off study.

***Pharmacokinetics.*** Serum $^{111}$In levels were drawn on day 1 at 5, 15, 30, 60, and 120 min. To determine whole-body clearance, four whole-body counts were obtained between days 2 and 6. Measurements taken on days of treatment occurred immediately before and 3 h after each infusion.

***Biodistribution.*** A minimum of two body images was obtained between days 1 and 6 and optionally on day 8. Single-photon emission computed tomography (CT) studies of the abdomen and pelvis and of suspected metastatic sites were used for all patients to assess $^{111}$In retention in tumor and normal tissue.

***Immunogenicity.*** Human anti-J591 antibodies in the serum of patients were assayed on days 4 and 8 and any day between days 15 and 19 using surface-enhanced laser desorption/ionization mass spectrometry technology as described previously (6).

***Complement activation.*** C3 and C4 serum protein levels were tested before treatment on days 4 and 8 and then any day between days 15 and 19 during each cycle.

***Antitumor effects.*** CT, magnetic resonance imaging, and bone scintigraphy, where applicable, were done at baseline, week 6, and following the fourth infusion of antibody. WHO criteria were used to assess responses. Tumor markers were assessed according to the same schedule when appropriate.

### Biostatistical considerations

The primary end points of the trial were to determine the safety, biodistribution, and pharmacokinetic profile of the drug. Dose-limiting toxicity was defined as a first cycle, drug-related, grade 3 nonhematologic toxicity or a first cycle, drug-related grade 4 hematologic toxicity using the National Cancer Institute Common Toxicity Criteria version 2 during the initial cycle of treatment. The maximum tolerated dose was defined as the highest dose level with an observed incidence of dose-limiting toxicity in one of six patients.

## Results

### Patients

Twenty patients with melanoma and cancers of the breast, colon, liver, and kidney were treated. Their demographics are shown in Table 1; treatments are shown in Table 2. Four patients were treated in cohorts 2 and 6. In cohort 2, a patient experienced unrelated grade 3 pain with a tumor-related pathologic fracture after receiving one dose and was replaced as she was unable to complete protocol requirements and came off study too quickly to assess either the side effects or antitumor activity of the drug. In cohort 6, a patient who experienced complications from a percutaneous nephrostomy tube placement came off study after receiving one dose and was replaced for the same reasons. The median number of cycles patients received was 2 (range, 1-4).

### Pharmacokinetics

The serum half-life ($t_{1/2}$), $C_{max}$, and area under the curve are shown in Table 3. The $t_{1/2}$ for each antibody mass was calculated from the serum radioactivity clearance curves shown in Fig. 1A, which are expressed as percentage injected dose per liter of serum. The 60 mg curve seems to be higher than the 100 mg curve in the figure because the 60 mg cohort, for uncertain reasons, had higher initial concentrations of antibody in the serum (in terms of percentage injected dose

**Table 1.** Patient characteristics

| Characteristic | No. patients ($N$ = 20) |
|---|---|
| Disease (primaries) | |
| Renal cell | 5 |
| Melanoma | 5 |
| Breast | 1 |
| Colon | 2 |
| Gastric | 1 |
| Hepatocellular | 1 |
| TCC renal pelvis | 2 |
| TCC bladder | 1 |
| Head and neck | 2 |
| Anatomic distribution of disease | |
| Bone | 3 |
| Lung | 13 |
| Lymph nodes | 10 |
| Liver | 6 |
| Kidney | 3 |
| Adrenal | 2 |
| Bladder | 1 |
| Other | 5 |
| Age | |
| Median | 65 |
| Range | 39-80 |
| Sex | |
| Male | 12 |
| Female | 8 |

Abbreviation: TCC, transitional cell cancer.

**Table 2.** J591 treatment schema

| Cohort | Unlabeled hu-J591 (mg) | $^{111}$In-labeled hu-J591 (on 2 mg), mCi | Total dose administered (mg) | Patients treated (*n*) | Median no. doses |
|---|---|---|---|---|---|
| 1 | 3 | 10 | 5 | 3 | 4 |
| 2 | 8 | 10 | 10 | 4 | 2 |
| 3 | 18 | 10 | 20 | 3 | 2 |
| 4 | 38 | 10 | 40 | 3 | 2 |
| 5 | 58 | 10 | 60 | 3 | 2 |
| 6 | 98 | 10 | 100 | 4 | 2 |

per liter). Nonetheless, the antibody cleared from the serum such that higher doses were associated with slower serum clearances. The most rapid serum clearance occurred at the 5 mg dose level, with a $t_{1/2}$ of 0.71 days. At doses of 10, 20, 40, 60, and 100 mg, the $t_{1/2}$ was 0.84, 1.86, 1.83, 3.32, and 3.56 days, respectively. These decreasing serum clearance rates resulted in commensurate stepwise increases in areas under the curve as well.

### Biodistribution

The antibody localized well to both bone and soft tissue sites of disease. Antibody images and standard scans were available on 18 patients. Representative scans are shown in Fig. 2. All patients showed tumor localization on antibody studies in at least one site of disease (bone or soft tissue) visualized by standard imaging studies. There seemed to be increasing uptake in tumor, with better delineation of lesions, at the higher mass amounts of protein; this is reflected particularly in higher tumor to nontumor and tumor to liver ratios, although the latter is also a function of decreasing liver uptake.

***Soft tissue disease.*** Eighteen patients had soft tissue disease as represented by metastases to lymph nodes, liver, skin, lung, or any other organ visualized on standard scans or physical exam. Seventeen (94%) of these patients showed demonstrable soft tissue uptake in at least one antibody scan. Organ-specific uptake was also examined. Nine of 13 (69%) patients who had lung disease on standard imaging also had lesions on antibody scans. Four of six (67%) patients with hepatic lesions visualized on CT scan showed uptake on antibody scans. In one instance each, the antibody scan was positive for hepatic, pulmonary, and nodal disease, which was not visualized on standard scans; similarly, five "false positives" were observed for other sites of soft tissue disease.

***Bone lesions.*** Six patients had bone disease on standard scans (this was calculated for bone scan and CT combined). All had positive antibody scans. Nine patients had negative standard scans and negative antibody scans. Three patients with negative standard scans had positive antibody scans.

***Hepatic uptake.*** Fractional hepatic uptake is shown in Fig. 1B. It seems that the liver saturates by the 60 mg dose, given the overlap between the 60 and 100 mg doses. There was no statistically significant difference between comparisons of fractional hepatic uptake on day 4 for the 5, 10, 20, and 40 mg cohorts. No difference was seen between the 60 and 100 mg cohorts, which were compared for day 5 given the paucity of data for these doses on day 4. There was a statistically significant decrease in fractional hepatic uptake between the lower doses. All patients in the 60 mg cohort had liver metastases, as did one patient each in the 40 and 20 mg cohorts; this may have resulted in a spurious increase in fractional hepatic uptake.

### Toxicity

Cycle 1 adverse events are shown in Table 4. No dose-limiting toxicity was seen nor were there any serious infusion-related events. Maximum tolerated dose was not reached. Of cycle 1 toxicities, only lymphopenia (25%, grade 3) and transaminitis (grade 1, in one patient) were felt to be possibly treatment related. In addition, one patient had grade 1 fever and one patient had grade 2 fever, which were felt possibly due to treatment. All other adverse events were felt to be due to illness or other medications. Maximum tolerated dose was not reached. Of all cycle adverse events, the only grade 3 event felt to be possibly drug related was also lymphopenia. No grade 4 events were seen. Treatment-related fatigue was seen in two patients, both of which were grade 1. Although there might be a theoretical concern for hepatotoxicity with repeated cycles, no such cumulative effects were seen, and all cycle treatment-related transaminitis was limited to one patient with a grade 1 transaminase elevation.

**Table 3.** J591 pharmacokinetics by dose level

| | 5 mg | 10 mg | 20 mg | 40 mg | 60 mg | 100 mg |
|---|---|---|---|---|---|---|
| $C_{max}$, mg/mL (SE) | 0.001 (3.874E-05) | 0.002 (7.349E-05) | 0.006 (1.302E-04) | 0.013 (4.117E-04) | 0.020 (2.723E-04) | 0.029 (2.347E-04) |
| AUC, mg · h/mL (SE) | 0.019 (2.153E-03) | 0.073 (1.222E-02) | 0.344 (6.805E-03) | 0.799 (3.650E-02) | 2.503 (1.770E-01) | 3.713 (8.690E-02) |
| Monoexponential $t_{1/2}$, days (SD) | 0.711 (7.30E-02) | 0.844 (2.07E-01) | 1.86 (3.07E-01) | 1.83 (3.27E-01) | 3.32 (1.87E-01) | 3.56 (2.34E-01) |

Abbreviation: AUC, area under the curve.

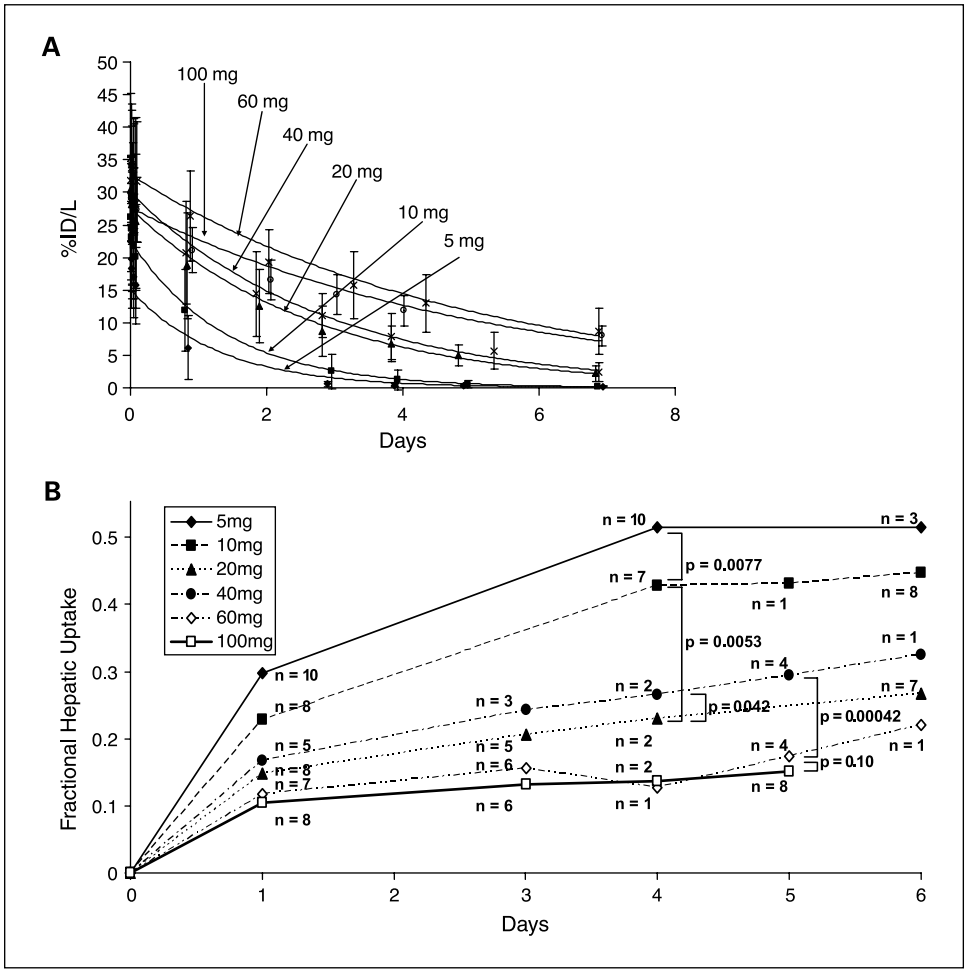


**Fig. 1.** *A,* clearance curves for J591 on a cohort by cohort basis, expressed as percentage injected dose per liter (*Y axis*) versus days (*X axis*). *B,* fractional hepatic uptake (*Y axis*) as plotted against time in days (*X axis*). *P* values were assigned to comparisons between cohorts where sufficient data permitted.

#### Antibody immunogenicity

No evidence of immunogenicity from the antibody was observed by pharmacokinetic data or by mass spectrometry.

#### Complement activation

Complement cascade activation was not apparent with treatment. Median C3 and C4 levels pretreatment, cycle 1 day 4, and cycle 1 week 3 levels were within laboratory normal limits.

#### Antitumor effects

Of the 18 patients evaluable for response and toxicity, 17 progressed after two to four cycles and 1 patient (renal) had stable disease after four cycles. No objective or clinical responses were observed. Three patients had tumor markers (carcinoembryonic antigen and CA19-9) followed in addition to radiographic studies. No significant posttreatment marker declines were seen.

## Discussion

This study examined the relationship between antibody mass, pharmacokinetic properties, biodistribution, and safety of J591 as a vascular targeting agent. Previously, we showed these relationships when using J591 as a means of targeting prostate cancer in patients with metastatic castration-resistant disease (5). In that prostate study, the cancer cell itself was the target. In this study, the target was tumor neovasculature. Because of the novelty of this approach and its distinction from targeting the cancer cell, we conducted a new study to show the pharmacokinetic properties, biodistribution, and safety of the antibody. This study achieved all of these aims.

Like many antibody treatments, the side effect profile will likely not determine the dose to be used in future phase I or II trials. Treatment was tolerated well at all dose levels, anti-J591 antibodies were not induced, complement was not activated, no dose-limiting toxicities were found, and maximum tolerated dose was not reached. Instead, the appropriate antibody mass will significantly be determined by pharmacokinetics and biodistribution. The rate of serum clearance in this study varied according to dose, with the fastest clearance rates associated with the lowest dose. In this study, the $t_{1/2}$ of the antibody was 0.711, 0.844, 1.86, 1.83, 3.32, and 3.56 days at doses of 5, 10, 20, 40, 60, and 100 mg of drug, respectively. Similarly, in our prostate cancer trial, the $t_{1/2}$ was 0.96, 1.9, 2.75, and 3.47 days for antibody masses of 10, 25, 50, and 100 mg (5).

The most likely reason for this relationship between dose and clearance is initial hepatic uptake of the antibody; after the liver saturates, serum clearance is longer. Although one could speculate that the liver is taking up the tracer and not the antibody, there is no reason to believe that $^{111}$In binds to PSMA (as it would more likely bind to transferrin), and therefore,

binding by the indium should not be affected by antibody mass. In our previous study, hepatic saturation seemed to occur by 25 mg. In this study, hepatic saturation seemed to occur at 60 mg, as fractional hepatic uptake did not differ between the 60 and 100 mg doses, suggesting that the liver binding sites were saturated. The fact that all of the patients in the 60 mg cohort had hepatic metastases adds credence to this hypothesized saturation point. Tumor sites in the liver should result in higher liver to blood count ratios, thereby increasing fractional hepatic uptake relative to the 100 mg dose, and should tend to separate the two curves by shifting the 60 mg curve "upward." Yet, in spite of these patients with hepatic metastases, the curves seem to overlap.

The J591 antibody was studied in a wide variety of solid tumors, and targeting to neovasculature was observed in the majority of patients. The recent approval of an agent that depletes circulating vascular endothelial growth factor has spurred interest in the development of agents that target the vascular components of solid tumors. By virtue of its targeting only to neovasculature and not to normal blood vessels, this antibody may have advantages compared with an agent that depletes vascular endothelial growth factor—tumor targeting will be more selective and lack of targeting to normal vessels may result in far fewer vascular side effects.

Selecting an appropriate dose for future trials will depend on the intended application of the antibody. We have previously shown that J591 induces antibody-dependent cell-mediated cytotoxicity, and the degree and duration of antibody-dependent cell-mediated cytotoxicity seem to be dose related. The antibody mass with the highest and most prolonged antibody-dependent cell-mediated cytotoxicity induction was 100 mg (5). In addition, we have previously shown using other antibodies that effective tumor targeting can only occur after hepatic saturation (13). If the same principles hold for vascular targeting using unlabeled antibody for immunotherapy, then the preferred antibody mass is one that saturates the liver, has immunologic activity, is economically feasible, and is safe. For the doses tested in this study, the 100 mg antibody mass meets these criteria.

Conceivably, higher doses may also be safe. Although this is the only study to use J591 at doses as high as 100 mg to target the neovasculature, studies of naked antibody using doses as high as 250 mg/m$^2$ have been tested in prostate cancer patients

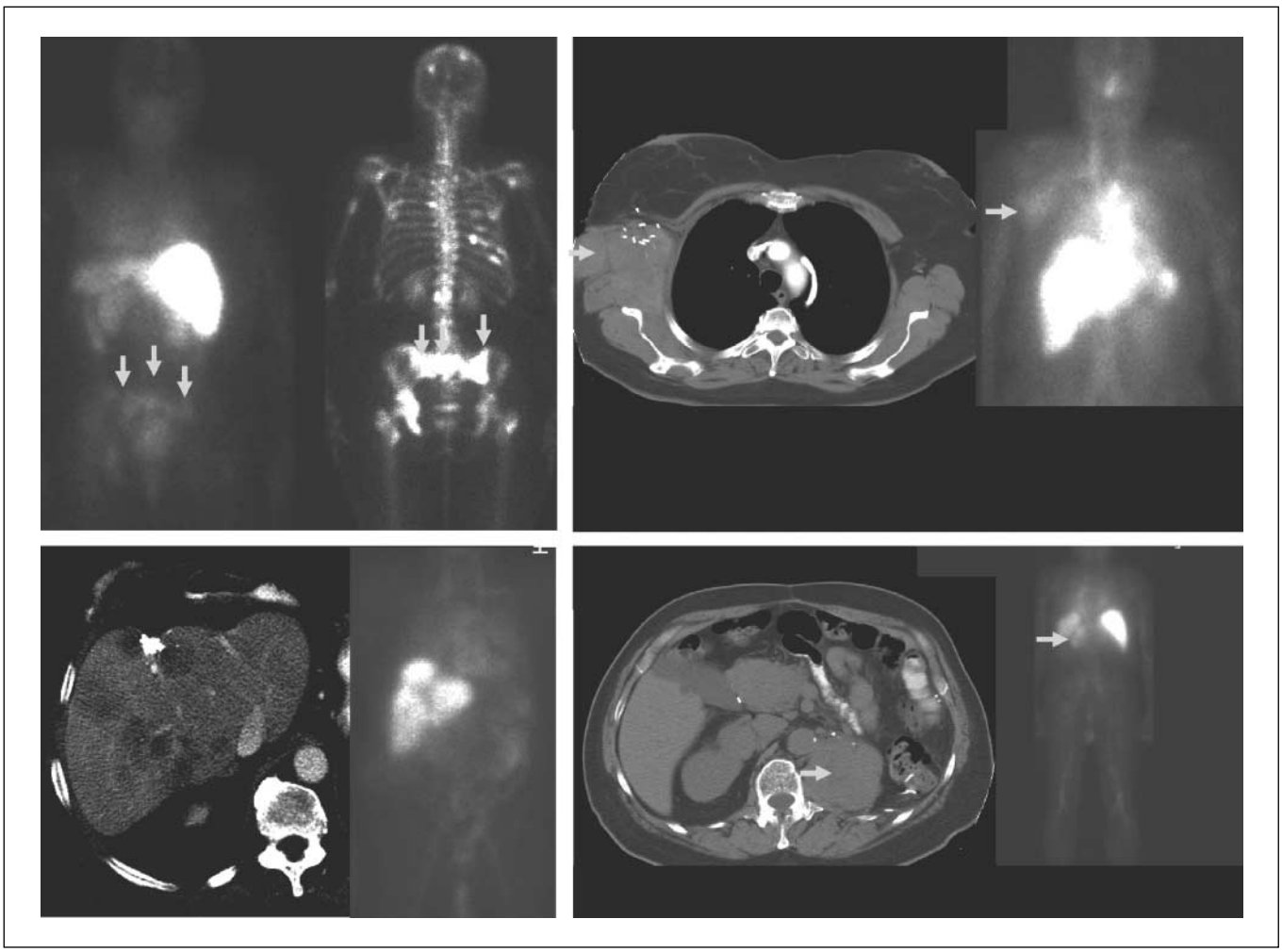

**Fig. 2.** Top left, patient with breast cancer and bone metastases who received 10 mg of J591 (*left picture*, J591 scan; *right picture*, bone scan). Top right, patient with melanoma and axillary nodal metastasis who received 10 mg of antibody (*left picture*, CT scan; *right picture*, J591 scan). Bottom left, patient with colon cancer who received 20 mg of antibody (*left picture*, CT scan; *right picture*, J591 scan). Note that the liver uptake in the patient with the hepatic uptake is dissimilar from the usual homogeneous hepatic uptake of patients with no liver metastases. Bottom right, patient with renal cell carcinoma who received 5 mg of antibody (*left picture*, CT scan; *right picture*, J591 scan, posterior view).

**Table 4.** Cycle 1 adverse events ($N$ = 20)

| Toxicity | Description | Toxicity grade | | | |
|---|---|---|---|---|---|
| | | 1, *n* (%) | 2, *n* (%) | 3, *n* (%) | 4, *n* (%) |
| Metabolic/hepatic | AST | 5 (25) | 3 (15) | 0 (0) | 0 (0) |
| | ALT | 4 (20) | 0 (0) | 0 (0) | 0 (0) |
| | Alkaline phosphatase | 6 (30) | 3 (15) | 0 (0) | 0 (0) |
| | Bilirubin | 3 (15) | 2 (10) | 1 (5) | 0 (0) |
| | Creatinine | 6 (30) | 1 (5) | 1 (5) | 0 (0) |
| | Hyperglycemia | 7 (35) | 6 (30) | 3 (15) | 0 (0) |
| | Hypoalbuminemia | 4 (20) | 2 (10) | 0 (0) | 0 (0) |
| | Hyponatremia | 3 (15) | 0 (0) | 1 (5) | 0 (0) |
| Hematologic | Hemoglobin | 6 (30) | 4 (20) | 2 (10) | 0 (0) |
| | Lymphopenia | 0 (0) | 0 (0) | 5 (25) | 0 (0) |
| Other | Fatigue | 7 (35) | 2 (10) | 0 (0) | 0 (0) |
| | Hemorrhage, other | 0 (0) | 0 (0) | 1 (5) | 0 (0) |
| | Infection with neutropenia | 0 (0) | 0 (0) | 1 (5) | 0 (0) |
| | Pain, other | 4 (20) | 1 (5) | 0 (0) | 0 (0) |
| | Rigors, chills | 9 (45) | 3 (15) | 0 (0) | 0 (0) |
| | Tumor pain | 1 (5) | 0 (0) | 1 (5) | 0 (0) |

NOTE: All grade 3 or 4 adverse events are shown, as are those grade 1 or 2 adverse events that affected >20% of patients.
Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase.

without toxicity (14). No study of unlabeled J591 has yet defined a maximum tolerated dose. Whether or not still higher doses would induce toxicity and whether higher doses would result in commensurately higher antibody-dependent cell-mediated cytotoxicity activity is something that would need to be established in future studies.

Dosing considerations for future radioimmunotherapy studies will have a different rationale. For radioimmunotherapy, it is important to ensure that antibody residence time in the blood be as low as possible to avoid irradiation of the marrow. At higher mass amounts of antibody, target saturation can result in prolongation of serum $t_{1/2}$ and thus of marrow radiation dose (15, 16). Therefore, a mass amount that saturates normal tissues and does not result in prolonged serum $t_{1/2}$ is optimal for radioimmunotherapy. An added advantage of a short serum $t_{1/2}$ is that fractionated radioimmunotherapy also becomes feasible (17). Although small antibody masses are associated with rapid serum clearance times, they also result in a high hepatic fractional uptake. Our data suggest that the antibody mass that mitigates both of these undesirable circumstances is 60 mg, at which the liver seems to be saturated with cold antibody and which has a relatively short $t_{1/2}$ of 3.3 days.

Other than issues of antibody mass, these data beg the question as to what diseases should be pursued in future antibody studies. Although no responses were seen in this study, this trial was not designed to detect responses as a primary end point. Indeed, unlabeled antibody might not induce tumor regression but rather inhibit future growth and prolong time to progression, particularly in patients with minimal disease burdens (18–21). Phase II trials will need to be designed with such nontraditional "responses" in mind.

In this phase I study, patients with melanoma and cancers of the breast, colon, liver, and kidney were treated. The antibody seemed to localize well to known sites of tumor involvement. Seventeen of 18 (94%) patients with soft tissue disease on standard scans showed uptake in the soft tissues on antibody scans as did 6 of 6 patients with bone disease. These data show selective targeting and good tumor localization. There was some heterogeneity when examining targeting on an organ-specific basis: the antibody scan localized to lung in 69% of patients with known lung disease and to liver in 67% of patients with known liver disease. However, these patients represent a wide range of solid tumors, some of which are highly dependent on vascular growth and others are not. In addition, vascularity may vary not just by the type of disease but by site of disease, as may PSMA expression. Further disease-specific studies are warranted. Candidate diseases for future study are kidney and colon cancers, where targeting angiogenic pathways has led to prolongation of patient survival and successful drug approval (22, 23). As this study shows the feasibility and tolerability of selectively targeting the neovasculature of nonprostate solid tumors by using an anti-PSMA antibodies, further studies of both unlabeled and radiolabeled J591 are justified.

## Acknowledgments

We thank Alyson Liedy, Maria Batraki, and Andrew Nagel for their assistance with data management and pharmacokinetic analysis.

## References

1. Israeli R, Powell CT, Corr JG, Fair WR, Heston WDW. Expression of the prostate-specific membrane antigen. Cancer Res 1994;54:1807–11.
2. Silver DA, Pellicer I, Fair WR, Heston WD, Cordon-Cardo C. Prostate-specific membrane antigen expression in normal and malignant human tissues. Clin Cancer Res 1997;3:81–5.
3. Carter RE, Feldman AR, Coyle JT. Prostate-specific membrane antigen is a hydrolase with substrate and pharmacologic characteristics of a neuropeptidase. Proc Natl Acad Sci U S A 1996;93:749–53.
4. Chengazi VU, Feneley MR, Ellison D, et al. Imaging

prostate cancer with technetium-99-7E11-5.3 (CYT-351). J Nucl Med 1997;38:675–82.
5. Morris MJ, Divgi CR, Pandit-Taskar N, et al. Pilot trial of unlabeled and indium-111-labeled anti-prostate-specific membrane antigen antibody J591 for castrate metastatic prostate cancer. Clin Cancer Res 2005;11:7454–61.
6. Milowsky MI, Nanus DM, Kostakoglu L, Vallabhajosula S, Goldsmith SJ, Bander NH. Phase I trial of yttrium-90-labeled anti-prostate-specific membrane antigen monoclonal antibody J591 for androgen-independent prostate cancer. J Clin Oncol 2004;22:2522–31.
7. Bander NH, Milowsky MI, Nanus DM, Kostakoglu L, Vallabhajosula S, Goldsmith SJ. Phase I trial of $^{177}$lutetium-labeled J591, a monoclonal antibody to prostate-specific membrane antigen, in patients with androgen-independent prostate cancer. J Clin Oncol 2005;23:4591–601.
8. Chang SS, Reuter VE, Heston WD, Bander NH, Grauer LS, Gaudin PB. Five different anti-prostate-specific membrane antigen (PSMA) antibodies confirm PSMA expression in tumor-associated neovasculature. Cancer Res 1999;59:3192–8.
9. Liu H, Moy P, Kim S, et al. Monoclonal antibodies to the extracellular domain of prostate-specific membrane antigen also react with tumor vascular endothelium. Cancer Res 1997;57:3629–34.
10. Conway RE, Petrovic N, Li Z, Heston W, Wu D, Shapiro LH. Prostate-specific membrane antigen regulates angiogenesis by modulating integrin signal transduction. Mol Cell Biol 2006;26:5310–24.
11. Chang SS, Reuter VE, Heston DWD, Bander NH, Grauer LS, Gaudin PB. Five different anti-prostate-specific membrane antigen (PSMA) antibodies confirm PSMA expression in tumor-associated neovasculature. Cancer Res 1999;59:3192–8.
12. Bander NH, Trabulsi EJ, Kostakoglu L, et al. Targeting metastatic prostate cancer with radiolabeled monoclonal antibody J591 to the extracellular domain of prostate specific membrane antigen. J Urol 2003;170:1717–21.
13. Divgi CR, Welt C, Kris M, et al. Phase I and imaging trial of indium-111 labeled anti-EGF receptor monoclonal antibody 225 in patients with squamous cell lung carcinoma. J Natl Cancer Inst 1991;83:97–104.
14. Nanus DM, Milowsky MI, Kostakoglu L, et al. Clinical use of monoclonal antibody HuJ591 therapy: targeting prostate specific membrane antigen. J Urol 2003;170:S84–8; discussion S8–9.
15. Sgouros G, Graham MC, Divgi CR, Larson SM, Scheinberg DA. Modeling and dosimetry of monoclonal antibody M195 (Anti-CD33) in acute myelogenous leukemia. J Nucl Med 1993;34:422–30.
16. Scheinberg DA, Lovett D, Divgi CR, et al. A phase I trial of monoclonal antibody M195 in acute myelogenous leukemia: specific bone marrow targeting and internalization of radionuclide. J Clin Oncol 1991;9:478–90.
17. Divgi CR, O'Donoghue JA, Welt S, et al. Phase I clinical trial with fractionated radioimmunotherapy using $^{131}$I-labeled chimeric G250 in metastatic renal cancer. J Nucl Med 2004;45:1412–21.
18. Zhang H, Zhang S, Cheung NK, Ragupathi G, Livingston PO. Antibodies against GD2 ganglioside can eradicate syngeneic cancer micrometastases. Cancer Res 1998;58:2844–9.
19. Nagy E, Berczi I, Sehon AH. Growth inhibition of murine mammary carcinoma by monoclonal IgE antibodies specific for the mammary tumor virus. Cancer Immunol Immunother 1991;34:63–9.
20. Eisenthal A, Lafreniere R, Lefor AT, Rosenberg SA. Effect of anti-B16 melanoma monoclonal antibody on established murine B16 melanoma liver metastases. Cancer Res 1987;47:2771–6.
21. Hara I, Takechi Y, Houghton AN. Implicating a role for immune recognition of self in tumor rejection: passive immunization against the brown locus protein. J Exp Med 1995;182:1609–14.
22. Hurwitz H, Fehrenbacher L, Novotny W, et al. Bevacizumab plus irinotecan, fluorouracil, and leucovorin for metastatic colorectal cancer. N Engl J Med 2004;350:2335–42.
23. Motzer RJ, Michaelson MD, Redman BG, et al. Activity of SU11248, a multitargeted inhibitor of vascular endothelial growth factor receptor and platelet-derived growth factor receptor, in patients with metastatic renal cell carcinoma. J Clin Oncol 2006;24:16–24.

Clinical Cancer Research

# Phase I Evaluation of J591 as a Vascular Targeting Agent in Progressive Solid Tumors

Michael J. Morris, Neeta Pandit-Taskar, Chaitanya R. Divgi, et al.

*Clin Cancer Res* 2007;13:2707-2713.