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Molecular Cancer Therapeutics

Spotlight on Clinical Response

A Pilot Clinical Study of Treatment Guided by Personalized Tumorgrafts in Patients with Advanced Cancer

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Abstract

Patients with many advanced solid cancers have very poor prognosis, and improvements in life expectancy are measured only in months. We have recently reported the remarkable clinical outcome of a patient with advanced, gemcitabine-resistant, pancreatic cancer who was later treated with DNA-damaging agents, on the basis of the observation of significant activity of this class of drugs against a personalized tumorgraft generated from the patient's surgically resected tumor. Here, we extend the approach to patients with other advanced cancers. Tumors resected from 14 patients with refractory advanced cancers were propagated in immunodeficient mice and treated with 63 drugs in 232 treatment regimens. An effective treatment regimen in the xenograft model was identified for 12 patients. One patient died before receiving treatment, and the remaining 11 patients received 17 prospectively guided treatments. Fifteen of these treatments resulted in durable partial remissions. In 2 subjects, no effective treatments were found. Overall, there was a remarkable correlation between drug activity in the model and clinical outcome, both in terms of resistance and sensitivity. The data support the use of the personalized tumorgraft model as a powerful investigational platform for therapeutic decision making and to efficiently guide cancer treatment in the clinic. *Mol Cancer Ther; 10(8); 1311–6. ©2011 AACR.*

Introduction

When it comes to anticancer drugs, a major obstacle is that one size does not always fit all (1). The individualization of cancer treatment may improve outcome and patient compliance (2). Although the rationale for this idea is strong and early clinical examples with targeted agents support this notion, the broad practical implementation of this concept remains difficult. In general, the field is mainly focused on finding the right patient for a given drug by implementing biomarkers predictive of drug action. For example, patients with lung cancer are now often assessed for mutations in the epidermal growth factor receptor (*EGFR*) gene because such genetic alterations confer susceptibility to inhibitors of the EGFR kinase (3, 4). Notwithstanding the importance of biomarker-driven approaches for cancer treatment, it has several challenges (5). First, it is a drug-centered rather than patient-centered approach, in which the main goal is to identify patients that may be good candidates for an agent. Second, these biomarkers often predict resistance rather than susceptibility (6). The frequency of most of these biomarkers is low within a given population and, thus, fails to provide a solution for most patients. Third, for multiple approved drugs, biomarkers are not known. Fourth, discoveries are, in general, restricted to diseases in which the drugs are approved, thereby limiting the possibility of finding effective applications in other tumor types. Finally, the positive predictive values are not perfect, and many patients, despite having the appropriate biomarkers, either do not respond or do so only transiently.

Personalized tumorgrafts developed in mice from patients' tumor tissues could potentially resolve some of the above-mentioned issues. These tumors recapitulate the biological characteristics of the disease of origin and are suitable for the quick assessment of the chemosensitivity of patients' cancer (7). Here, we used the tumorgraft model to personalize the treatment course for patients with advanced cancers.

Materials and Methods

Patients

Patients were enrolled in the Johns Hopkins University protocol J0507 (NCT00276744) or in the Hospital de

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Note: Supplementary data for this article are available at Molecular Cancer Therapeutics Online (http://mct.aacrjournals.org/).

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Table 1. Patient characteristics and outcome of treatments based on xenograft predictions

Identification number	Gender	Age (years)	Tumor	Xenograft	Prior systemic therapy	Systemic therapy administered on the basis of xenograft data	Response	Duration of response (months)
JH033	Male	63	Pancreatic ductal adenocarcinoma	Primary tumor	Gemcitabine	Mitomycin C, cisplatin	PR	50+
JH082	Female	64	Pancreatic ductal adenocarcinoma	Primary tumor	None	Gemcitabine	Progressive disease	
JH102	Female	66	Pancreatic ductal adenocarcinoma	Primary tumor	None	Gemcitabine	Stable disease	
JH161	Female	70	Pancreatic ductal adenocarcinoma	Primary tumor	None	Gemcitabine	Stable disease	
CBI-0601	Female	55	Leiomyosarcoma	Pelvic metastasis	Adriamycin, ifosfamide	Gemcitabine, docetaxel, bevacizumab	PR	9
					Gemcitabine, doxetaxel	Capcetabine, temozlomide	PR	9
CBI-0602	Male	45	Mesenchymalchondro sarcoma	Lung mestastasis	VAC	Docetaxel, irinotecan, bevacizumab	PR	9
					Sunitinib		Stable disease	4
CBI-0603	Male	50	Non–small cell lung carcinoma	Primary tumor	Carboplatin, paclitaxel	Sorafenib, irinotecan, bevacizumab	PR	9
					Pemetrexed		No response	
CBI-0701	Male	36	Myoepithelioma	Primary tumor	None	Doxorubicin, cisplatin	Progressive disease	
						Imatinib	Progressive disease	
						Carboplatin-bevacizumab	Progressive disease	
CBI-0803	Male	54	Esophageal adenocarcinoma	Liver metastasis	Epirubicin, 5-fluorouracil, cisplatin	Irinotecan-cetuximab-bevacizumab	PR	14
						Abraxane-avastin	PR	9
						Ixabepilone-avastin	PR	9
						Ixabepilone-navelbine	PR	6+
CBI-0805	Female	44	Colon adenocarcinoma	Liver metastasis	FOLFOX	Irinotecan	PR	14
						Capecitabine, lapatanib	PR	6+
CBI-0810	Female	36	Breast cancer	Liver metastasis	5-fluorouracil, cyclophosphamiirinotecan, bevacizumab	None	No response	
							No response	
CBI-0901	Male	49	Rhabdomyosarcoma	Lung metastasis	None	Docetaxel, gemcitabine, bevacizumab	PR	8
CBI-0907	Female	62	Colon	Liver metstasis	FOLFOX	Cetuximab, irinotecan	PR	6
					Irinotecan		PR	15
CBI-0914	Male	58	Melanoma	Lung metastasis	None	B-raf inhibitor	PR	6

Note: When tumorgrafts reached approximately 150 mm^3, animals were randomized (5 mice with tumors on both flanks per group) and dosing initiated. Details on drugs, doses, and schedules are provided in Supplementary Table S1.

Abbreviations: VAC, vinblastine, doxorubicin and cisplatin; FOLFOX, folinic acid, 5-fluorouracil, oxaliplatin.

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Madrid protocol FHM.06.10. In these studies, patients with refractory solid tumors or early stage, poor prognosis cancers had the opportunity to have their tumors implanted in nude mice. A fresh tumor specimen was collected, either at the time of surgical resection or by a tumor biopsy, and implanted in immunodeficient mice and propagated (8, 9).

Preclinical studies

When tumorgrafts reached approximately 150 mm^3, animals were randomized (5 mice with tumors on both flanks per group) and dosing was initiated (Supplementary Table S1). Final tumor volumes were compared using a 2-tailed ANOVA, adjusted for multiple comparisons. A rank list of effective treatments was provided to the treating physician, who then selected the patient treatment.

Biological and pharmacologic studies

Gene expression analysis was done using Affymetrix U 133 Plus 2.0 gene arrays (10). Gene set analysis was done using the GSEA software V2.0.2. Genes represented by more than one probe were collapsed using the Collapse Probes utility to the probe with the maximum value. We used unsupervised clustering analysis to classify responders and resistant tumors on the basis of the expression of irinotecan pathway genes. The intratumor concentrations of irinotecan (CPT 11) and the metabolite SN38 were measured in tumors collected 6 hours after the last dose of CPT11, as previously described (11).

Results

A total of 14 tumorgrafts were obtained from 14 patients from either primary resected tumor (6 patients) or resected metastasis (8 patients) and were treated with 63 different anticancer agents spanning 33 unique mechanisms of action in 232 single-agent or combination treatments. Supplementary Table S1 provides details about drugs, mechanism of action, combinations, dose and schedules, and activity noted in the mouse model. A regimen was considered active if it resulted in a tumor growth inhibition (TGI) $\geq$80% and/or a partial response (PR) rate $\geq$50%. In 2 tumors, JH082 (pancreatic cancer) and CBI-0701 (myoepithelioma of the salivary gland), no effective regimen was found in the 4 and 13 treatments tested, respectively.

Table 1 depicts the most relevant patient characteristics. Three patients with standard-of-care–resistant metastatic cancers remain alive at 50+, 38+, and 20+ months.

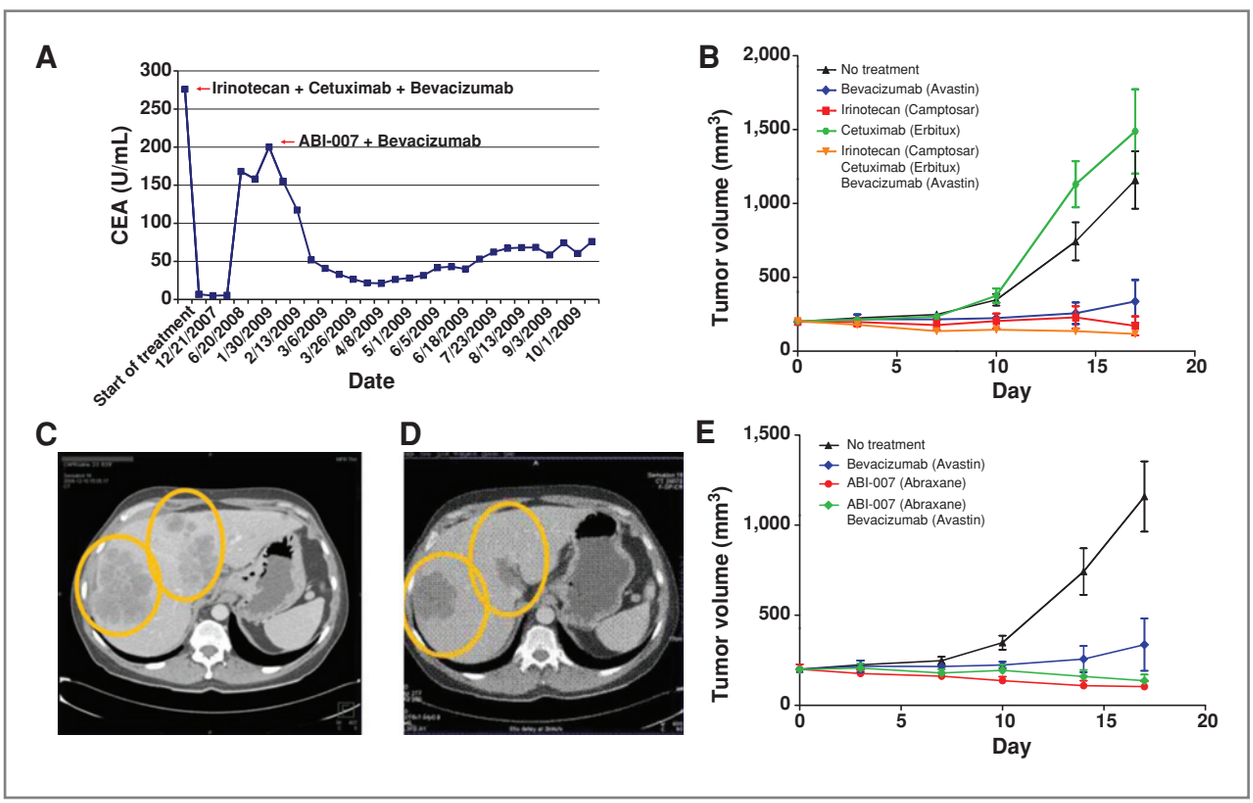

Figure 1. Clinical outcome of patient CBI-0803 and response to anticancer agents in patient's xenografts. A, time course of CEA levels. B, tumor growth curve of patient's personalized tumorgraft, treated with the indicated agents at the dose and schedules. C and D, CT scan of the abdomen, before and after treatment with irinotecan-cetuximab and bevacizumab, showing a marked decrease in tumor volume. E, tumor growth curve of patient's tumor treated with the indicated agents.

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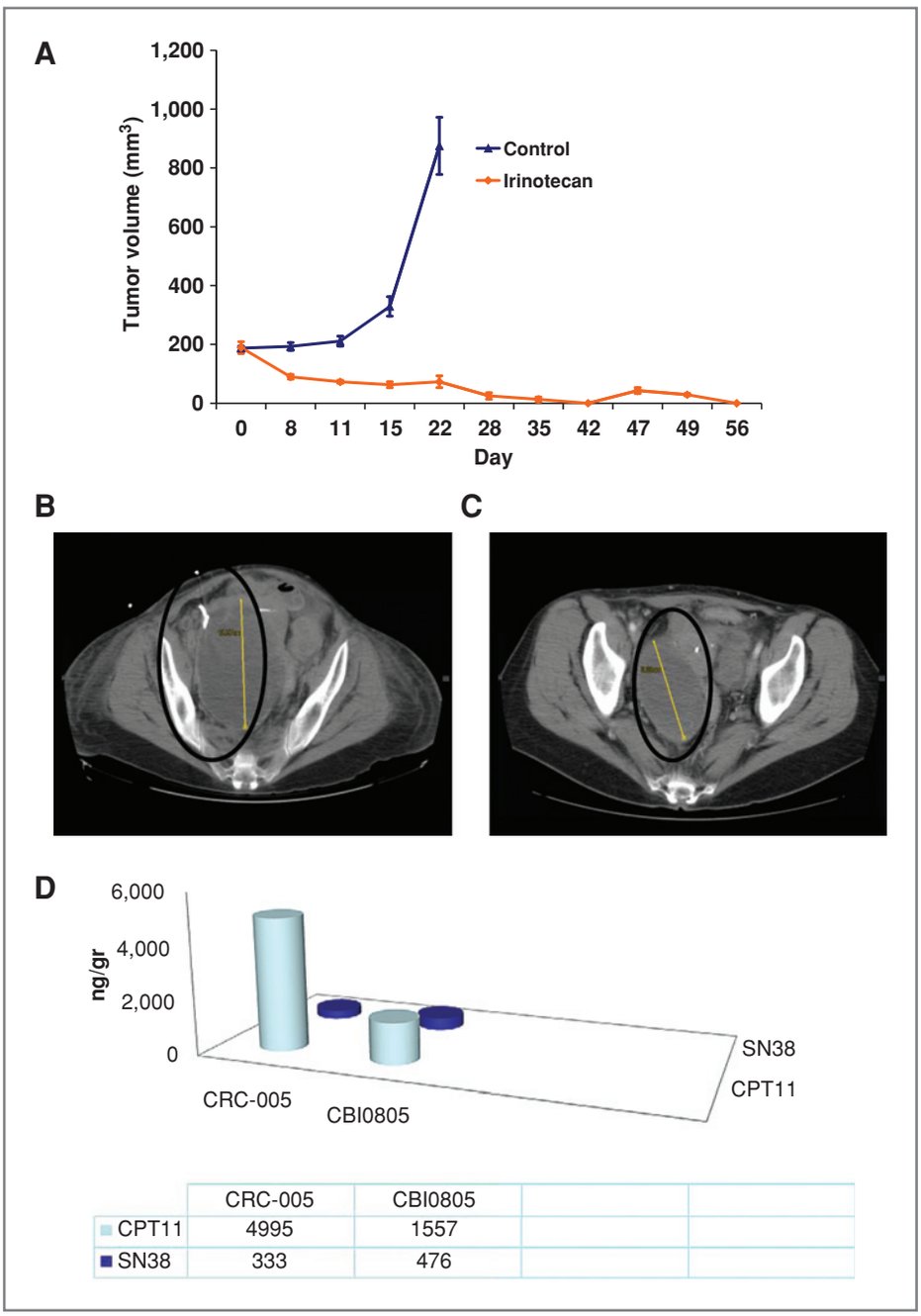
Patient CBI-0803 is a 54-year-old male who presented with stage IV gastroesophageal adenocarcinoma with liver and lung metastasis. The patient was initially treated with an epirubicin-cisplatin-capecitabine regimen with a PR that lasted 8 months. Subsequently, disease progression developed with lung and liver metastasis and an elevation of the carcinoembryonic antigen (CEA) tumor marker (Fig. 1A). At that point, a tumorgraft generated from a resected liver metastasis had been treated with 17 different drugs in 35 combinations. As shown in Fig. 1B, the tumorgraft responded to the combination of irinotecan, bevacizumab, and cetuximab, which was recommended for clinical use. With this treatment, the patient achieved a PR in the liver metastasis (Fig. 1C, pretreatment, and D) that lasted 14 months. At that point, his CEA started to increase again to 200 UI/mL. Data from his personalized tumorgraft indicated susceptibility to nab-paclitaxel (ABI-007) in combination with several angiogenesis inhibitors (Fig. 1E). The patient received treatment with nab-paclitaxel in combination with



	CRC-005	CBI0805		
CPT11	4995	1557		
SN38	333	476		

Figure 2. Clinical outcome of patient CBI-0805 and remarkable antitumor potential of irinotecan in patient's xenografts. A, response of patient's tumorgraft to single-agent irinotecan illustrates a complete eradication of the tumor. B, CT scan of pelvis before treatment. C, CT scan of pelvis showing a significant reduction in a metastatic pelvic mass. D, intratumor concentration of irinotecan and SN38 in this patient's xenograft and an irinotecan-resistant colorectal cancer (CRC) tumorgraft (CRC-005), showing heightened retention of SN38 in CBI-0805.

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bevacizumab, with a normalization in CEA levels that has been maintained for 8 months (Fig. 1A).

The second patient, CBI-0805, is a 44-year-old female diagnosed with stage III colon cancer. The patient was treated with surgery and 5-fluorouracil–irinotecan chemotherapy. After 2 years, she presented with liver metastasis and underwent tumor resection. Tumorgraft from this patient was extremely sensitive to irinotecan (Fig. 2A). Six months after surgery, the patient progressed with a large pelvic mass (Fig. 2B, pretreatment, and C) that caused severe pain and hydronephrosis requiring nephrostomy tubes. She was treated with single-agent irinotecan and achieved a PR with resolution of her pain and restoration of urinary flow, which lasted 14 months.

The third case is a 61-year-old male who underwent a distal pancreatectomy for a pT3N1M0 ductal adenocarcinoma of the pancreas. The clinical outcome of this patient has been recently reported (12), and the patient remains disease free 50+ months after diagnosis.

Tumor CBI-0805 showed remarkable sensitivity to irinotecan. To explore the potential mechanisms of sensitivity, we compared this tumor gene expression profile with that of 4 additional colorectal cancer tumorgrafts with known response to irinotecan. Using a previously published irinotecan 24-gene expression signature, the 3 irinotecan-sensitive tumors cluster together (Supplementary Fig. S1; ref. 11). A closer analysis of the expression of the candidate genes in this case shows that this patient tumor is characterized by high expression of the irinotecan membrane transporters ABCB1 and ABCG2 and low expression of the genes involved in the catabolism of SN38, the active metabolite of irinotecan (CYP3A4 and 5 and UGT1A1). Consistently with this gene expression profile, CBI-0805 had a higher concentration of SN38, which represented 30% of the parental drug irinotecan (CPT11; Fig. 2D). This finding suggests that this cancer's unique sensitivity to irinotecan is based on its ability to retain high concentrations of the active metabolite SN38.

Discussion

This report summarizes the results of a pilot study in patients with advanced cancer whose treatments were selected on the basis of activity against a personalized tumorgraft developed from the patient's own cancer. The data show a remarkable correlation between drug activity in the model and clinical outcome, both in terms of resistance and sensitivity. The treatments selected for each individual patient were not obvious and would not have been the first choice for a conventional second- or third-line treatment. This observation is perhaps best illustrated by the combination of cetuximab and bevacizumab in patient CBI-0803, as the combination of these 2 agents is not recommended in clinical practice (13). The objective response rate was 88% for treatments deemed effective by the model and tested in the patients. Overall, 11 of 14 patients achieved a PR. The expected response rate with phase I agents, the only available option for some of these subjects, is less than 10% (14). This preclinical-clinical correlation supports the value of personalized tumorgraft models as being predictive of clinical outcome. The abundant tumor materials obtained by propagating the cancer in mice allow biological and pharmacologic studies in the validated models to understand the observed effects. This process led, as recently reported, to the discovery of PALB2 mutations in a mitomycin C–responsive patient (12, 15).

The limitations to this approach certainly challenge the broad clinical application of the process and will need to be resolved before this can be first tested in a randomized clinical trial. The process requires large amounts of fresh tumor material and intense resources to generate the tumorgraft. Even in the best conditions, 25 to 30% of implants fail, and those that engraft require 6 to 8 months of additional propagation to be useful for treatment.

In summary, this work shows that personalized tumorgrafts can be used to individualize patient treatment and to discover determinants of drug response. In those patients in whom an effective treatment is found, the clinical activity is remarkable. Nevertheless, this process has limitations in efficiency, speed, and cost, but given the promising response data, additional investigation to solve these issues is warranted.

Disclosure of Potential Conflicts of Interest

D. Sidransky is a consultant to Champions Biotechnology, Inc. (CBI) and chairman of the company's board of directors. The terms of this arrangement are being managed by the Johns Hopkins University in accordance with its conflict of interest policies. M. Hidalgo and J. Martell are consultants and stock holders in CBI. E. Bruckheimer and S. Strawn are employees of CBI. The other authors declare no potential conflicts of interest.

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