Transhepatic Arterial Chemoembolization with Oxaliplatin-eluting Microspheres (OEM-TACE) for Unresectable Hepatic Tumors

GUIDO POGGI[1], PIETRO QUARETTI[2], CLAUDIO MINOIA[3], GIOVANNI BERNARDO[1], MARIO REGAZZI BONORA[4], RAFFAELLA GAGGERI[3], ANNA RONCHI[3], CESARE MASSA SALUZZO[2], ANDREA AZZARETTI[2], GIUSEPPE RODOLICO[2], MICHELA MONTAGNA[4], ALESSIO AMATU[1], CRISTINA TERAGNI[1], ILARIA PALUMBO[1], ELENA TRAVERSO[1], STEFANO TONINI[1], LAURA VILLANI[5], MARIO SCELSI[5], PAOLA BAIARDI[6], MARIA GRAZIA FELISI[6], FEDERICO SOTTOTETTI[1], BARBARA TAGLIAFERRI[1] and ALBERTO RICCARDI[1]

Departments of [1]Oncology and [5]Pathology, [3]Laboratory for Enviromental and Toxicological Testing, and [6]Consortium for Biological and Pharmacological Evaluations, University of Pavia, IRCCS Fondazione S. Maugeri, Istituto Scientifico di Pavia; [2]Unit of Interventional Radiology, [4]Clinical Pharmacokinetic Unit, IRCCS Policlinico San Matteo, Pavia, Italy

Abstract. *Background: While conventional transhepatic arterial chemoembolization (TACE) is accepted worldwide as an effective treatment for patients with unresectable hepatocellular carcinoma (HCC), its use in other hepatic tumors is not supported by randomized studies. Preliminary results have shown that new drug-eluting microspheres (DEM) seem to optimize TACE procedures. The aim of this study was to evaluate the capability of HepaSphere™ to load oxaliplatin and their pharmacokinetic outcome. The feasibility and safety of treatment with oxaliplatin-eluting microspheres (OEM-TACE) was also evaluated in patients with unresectable liver metastasis of colorectal cancer and unresectable intrahepatic cholangiocarcinoma. Patients and Methods: An inductively coupled plasma mass spectrometer (ICP-MS) was used to quantify the oxaliplatin bound to microspheres and the oxaliplatin in liver biopsies. Fifteen patients (8 with colorectal carcinoma liver metastases, 7 with intrahepatic cholangiocarcinoma) were treated with 27 sessions of OEM-TACE. Results: The data suggested that t he microspheres can bind oxaliplatin entirely. The pharmacokinetic parameters were significantly different between the OEM-TACE patients and a control group of patients treated with oxaliplatin chemotherapy. The mean oxaliplatin concentration within the tumor was twenty-times higher than the extratumoral liver concentration in the OEM-TACE patients. According to response evaluating criteria in solid tumors (RECIST), stable disease was observed in 8 out of the 15 patients (53.3%), a partial response in 2 (13.3%) and intrahepatic or extrahepatic tumor progression in 5 out of the 15 patients (33.3%). No major adverse event (AE G3/4) occurred. Conclusion: TACE with oxaliplatin-loaded microspheres is a safe and feasible treatment without major adverse events and with a favorable pharmacokinetic profile.*

Correspondence to: G. Poggi, MD, U.O. Oncologia II, Fondazione Maugeri 27100 Pavia, Italy. Tel: +39 0382 592675, Fax: +39 0382 592206, e-mail: guido.poggi@fsm.it

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Transhepatic arterial chemoembolization (TACE) is generally considered the mainstay of therapy for patients with intermediate hepatocellular carcinoma (HCC) and adequate preservation of liver function as demonstrated by two recent randomized trials and two meta-analyses (1-4), but fewer results are available for the treatment of hepatic metastases (5) in hepatic colorectal metastasis or cholangiocarcinoma. Although not supported by well-designed randomized studies, nevertheless phase II studies of chemoembolization for metastatic colorectal cancer have reported promising results in patients with good performance status and disease isolated to the liver (6-9) and impressive outcomes have been reported by Buerger *et al.* in patients with unresectable cholangiocarcinoma (10). Although there is no one standardized accepted protocol, typically conventional TACE consists of a selective embolization of the tumor-feeding arteries with an emulsion of a chemotherapeutic agent, the most used being doxorubicin, cisplatin and mitomycin, combined with an ethiodized oil (Lipiodol; Savage Laboratories, Melville, NY, USA) whose role is to emulsify the drugs and carry them into the lesions.

Embolization particles or Gelfoam (Pharmacia and Upjohn, Kalamazoo, MI, USA) are then used to reduce arterial inflow, induce tissue ischemia, and decrease the washout of the chemotherapeutic agents into the systemic circulation in order to prolong the contact time between cancer cells and drugs. The ultimate goal of TACE is to obtain the highest sustained concentration of drugs within the tumor, with the lowest concentration of the drug in peripheral blood.

Unfortunately, the process of emulsification between lipiodol and drugs is very unstable and the two components begin to separate as soon as they are injected into the hepatic arterial circulation so that the pharmacokinetic profiles of drugs are similar whether they are injected as an intra-arterial infusion or introduced with the conventional TACE protocol (11). To optimize the drug delivery to the tumor, two kinds of new drug-eluting microspheres (DEM) are now available: polyvinyl alcohol-based microspheres (DC Bead™: Biocompatibles UK Ltd., Farnham, UK) and superabsorbent polymer microspheres (HepaSphere™: Biosphere Medical, Roissy CDG Cedex, France). Such microspheres can be loaded with chemotherapeutic agents and delivered directly into the tumor, achieving high intratumoral concentration and low plasma concentrations. The capability of these microspheres to be loaded with several types of chemotherapeutic agents provides a new tool for delivering a high dose of effective drugs in tumors with different histotypes or decreasing plasma levels of drug and for limiting systemic side-effects.

Oxaliplatin is a platinum coordination complex widely used in the treatment of metastastic colorectal cancer (CRC) in combination with fluoropyrimidines (12) and more recently its use has been extended to the treatment of cholangiocarcinoma in combination with gemcitabine (13). In this study the capability of HepaSphere™ to load oxaliplatin and the pharmacokinetic outcomes of this new oxaliplatin delivery system were evaluated. The feasibility and safety of the treatment with oxaliplatin-eluting microspheres TACE (OEM-TACE) was also evaluated in patients with unresectable liver metastasis of CRC and in patients with unresectable intrahepatic cholangiocarcinoma.

Patients and Methods

Patients. Between July 2006 and December 2006 fifteen patients with unresectable liver tumor were enrolled to be treated with OEM-TACE. A total of twenty-seven OEM-TACE procedures were performed. Each patient received one treatment cycle consisting of one to three sessions of TACE based on liver tumor involvement. Seven to fifteen days after OEM-TACE, 11 of the patients were also treated with 16 sessions of percutaneous radiofrequency thermal ablation (RFTA). The patient population comprised nine males and six females (51-83 years old; mean 64.4 years; SD 8.087 years; median 63 years). Five patients had metachronous and three patients had synchronous CRC liver metastases and seven patients had intrahepatic cholangiocarcinoma. Four patients (3 males and 1 female) with metachronous CRC hepatic metastases treated with oxaliplatin-containing systemic chemotherapy (FOLFOX 4) were considered the control group. Table I shows the characteristics of the patients. The patients were not considered for surgical resections due to age, prior hepatic metastectomy, disease extent, comorbidity or refusal to consent to surgery. All the patients had previously undergone one or more lines of chemotherapy. Exclusion criteria included evidence of extrahepatic disease, renal failure, hepatic failure (bilirubin >2 mg/dl; serum albumin less than 35 g/l), impaired clotting test (platelet count below 50,000 mm^3 or prothrombin activity below 50%), white blood cell count <3,000/mm^3, evidence of portal thrombosis and symptomatic peripheral neuropathy. Written informed consent was obtained from all the patients prior to treatment. Proof of malignancy of at least one hepatic lesion was obtained in all the patients by percutaneous core-needle biopsy.

Pre-clinical study. A pre-clinical study *in vitro* was carried out to evaluate if the microspheres (HepaSphere™) were capable of being loaded with oxaliplatin before their administration in the TACE procedure. The determination of inorganic platinum in the oxaliplatin-binding microspheres at different time-points was performed with an inductive coupled plasma mass spectrometer (ICP-MS). A 50 mg vial of HepaSphere™ was preloaded with different levels of oxaliplatin and then diluted, after mixing it for 5, 15, 30 or 45 min, in contrast medium (iodixanol, Visipaque® 270; Amersham Health, Milan, Italy). The highest dose of drug tested was 400 mg oxaliplatin/50 mg HepaSphere™. The solution obtained was passed through a filter (GHP Acrodisc, 0.45 μm; PALL Life Science, VWR International srl, Milano) and then diluted (1:10 v/v) in aqueous nitric acid solution (1% v/v) before analysis.

Determination of platinum in biological samples. The determination of platinum in biological matrices was carried out by ICP-MS using an ELAN DRC II instrument (Perkin-Elmer Sciex Instrument, Concorde, Ontario, Canada), equipped with a cyclonic spray chamber and a Meinhard type concentric nebulizer. The instrument operated in standard mode and the selected analyte was ^{195}Pt. In order to eliminate the matrix effect, ^{193}Ir, an isotope with chemical-physical properties similar to those of Pt, was used as internal standard (IS). Standard solutions of Pt for calibration curves and of Ir as IS were supplied by CPI International (Amsterdam, the Nederlands). Ultrapure 65% HNO_3 was provided by Merck (Darmstadt, Germany), and the high-purity water used throughout was prepared by a Milli-Ro, Milli-Q system (Millipore, Bedford, MA, USA). Oxaliplatin was evaluated by ICP-MS in plasma, urine, ultrafiltrate and liver biopsies. After collection, 500 μl plasma samples were immediately ultra-filtrated centripetally in apposite centrifugal devices with 10 kDa molecular weight cut-off membrane (Nanosep Centrifugal Devices, Pall Life Sciences) at 2,000 rpm for 8 minutes to obtain the ultrafiltrate. Plasma and ultrafiltrate were diluted in polypropylene tubes, with 1% HNO_3 (v/v) (Suprapur, Merck) at a rate of 1:100 and 1:30 (v/v), respectively. The urine specimens were diluted 1:100 (v/v) with 1% HNO_3 (v/v) (Suprapur, Merck). The liver biopsy samples were mineralized in a microwave oven (MarsXpress, CEM Corporation, Matthews, North Carolina) with HNO_3 at 65% and then diluted (1:40, v/v) with aqueous nitric acid solution (1% v/v).

OEM-TACE procedure. A 50 mg vial of HepaSphere™ with a diameter ranging from 50 to 100 μm was preloaded by mixing it for at least 10-15 min with 50 mg oxaliplatin (Eloxatin, Sanofi-

Table I. *Characteristics of patients treated with OEM-TACE (see text for details). Controls were treated with Folfox4.*

Patient N°	Age/ Gender	ECOG	Primary tumor	Metastasis	N° Lesions	Treatment before TACE	OEM-TACE session	Dose oxaliplatin (mg)	RFTA N°	Response (RECIST)
1	64/Female	0	CRC	Synchronous	3	FOLFIRI	3	25/ 50/50	2	PR
2	66/Female	0	Cholangio	-	2	GEMOX	3	45/50/92	2	PR
3	59/Female	1	Cholangio	-	2	GEMOX	2	50/26	-	P
4	74/Female	1	Cholangio	-	1	GEMOX	1	35	1	SD
5	83/Male	1	Cholangio	-	1	GEMOX	2	50/50	-	SD
6	71/Male	0	Cholangio	-	>3	GEMOX	1	100	2	SD
7	71/Male	0	Cholangio	-	>3	GEMOX	1	61	1	P
8	62/Female	1	CRC	Metachronous	>3	FOLFOX	3	50/36/40	-	P
9	54/Male	0	CRC	Synchronous	>3	FOLFOX	2	50/50	2	SD
10	51/Female	0	CRC	Metachronous	2	FOLFOX FOLFIRI	2	28/50	1	SD
11	63/Male	0	CRC	Metachronous	2	FOLFOX FOLFIRI	1	50	2	SD
12	60/Male	0	CRC	Metachronous	>3	FOLFOX FOLFIRI	2	50/50	1	P
13	60/Male	0	CRC	Metachronous	2	FOLFOX FOLFIRI	1	50	1	P
14	58/Male	0	Cholangio	-	1	GEMOX	2	50/50	-	SD
15	71/Male	0	CRC	Synchronous	1	-	1	50	1	SD
Control 1	64/Male	0	CRC	Metachronous	>3	-	-	135	1	-
Control 2	72/Female	1	CRC	Metachronous	>3	-	-	140	1	-
Control 3	58/Male	0	CRC	Metachronous	3	-	-	125	1	-
Control 4	66/Male	1	CRC	Metachronous	2	-	-	135	1	-

ECOG: Eastern Cooperative Oncology Group, performance status, RFTA: radiofrequency thermal ablation, CRC: colorectal carcinoma, Cholangio: intrahepatic cholangiocarcinoma, FOLFIRI: 5-fluorouracil, leucovorin, irinotecan, GEMOX: gemcitabine, oxaliplatin, FOLFOX: 5-fluorouracil, leucovorin, oxaliplatin, RECIST: response evaluation in solid tumors, PR: partial response, P: progression; SD: stable disease.

Aventis) and diluted with 5 ml of the non-ionic contrast medium (Visipaque®). The particles were then further diluted with the same contrast medium to obtain a 30 ml solution and continuously mixed with a stopcock between two 20 ml syringes before being injected through a 1 ml Luer-Lock syringe. Antibiotic prophylaxis was given before the procedure (cefotaxime 2 g *i.v.*) A femoral approach was obtained through a 25 cm long 5 or 6 F introducer in order to straighten sharp bending of elongated iliac arteries. Baseline selective angiography of the celiac trunk, common hepatic and mesenteric arteries was performed with a proper shaped tip catheter prolonging the time of fluorography to obtain diagnostic imaging of the portal system. In case (one patient) of obstruction of the celiac trunk, highly selective angiography of the hepatic artery was achieved by negotiating a 1.7 F microcatheter (Echelon; Micro Therapeutics, Irvine, CA, USA), through the pancreatic-duodenal arcade stabilizing the coaxial system with a 6 F guiding catheter in the superior mesenteric artery. The particles were injected manually under continuous fluoroscopy assessment with a Luer-Lock syringe through the microcatheter positioned as distal as possible into the right or left hepatic artery, avoiding any dangerous reflux inside the arteries feeding the gallbladder or the gastric wall. The angiographic end-point was achieved when stagnant flow in the pathological area was noted.

Patient monitoring and response criteria. Serum liver transaminases, total bilirubin, alkaline phosphatase, complete blood count and platelet count, serum amylase and lipase were monitored for each patient just before and 24 h after each TACE procedure. Carcinoembryogenic antigen and cancer antigen 19.9 were measured on study entry, before each TACE procedure and then every month until the patient was off the study. Restaging with abdominal CT scan and chest radiograph was carried out just before TACE, 1 month after the last TACE and then every 3 months until progression. All the patients were followed-up until death for progression-free survival (PFS) and overall survival (OS). Assessment of response was evaluated by response evaluation criteria in solid tumors (RECIST) (14).

Determination of intratumoral and extratumoral concentrations of oxaliplatin. After the TACE or after the end of systemic chemotherapy a percutaneous ultrasound-guided core-needle biopsy of the lesion and of normal hepatic perilesional tissue were obtained in eleven patients (7 from the OEM-TACE group and 4 from the systemic chemotherapy group) to determine both intratumoral and extratumoral concentrations of oxaliplatin.

Oxaliplatin pharmacokinetics. Oxaliplatin pharmacokinetics were recorded in twelve patients at the first procedure of OEM-TACE and

in three of them in the second procedure for a total of fifteen determinations. Oxaliplatin pharmacokinetics were also performed in three of the patients with CRC hepatic metastases from the control group. Peripheral blood samples to evaluate oxaliplatin levels in the plasma and ultrafiltrate were collected at baseline and at 30 min, 1, 2, 4, 8, 12, 24, 48 h and 3, 4, 5, 6, 7 days after the OEM-TACE procedure. The peak oxaliplatin concentrations (C_{max}) and the time taken to attain these levels (T_{max}) were determined, along with the area under the curve (AUC). Urine samples were obtained at baseline and at every day until the seventh day. Concentration *versus* time curves were constructed over seven days. Oxaliplatin concentration–time data were analyzed using a statistical pharmacokinetics population program (P-Pharm, Version 3; Simed, Creteil, France). P-Pharm uses a maximum posterior probability (MAP) Bayesian fitting procedure to combine knowledge and the available individual information to estimate the individual parameters and compute approximate statistical tests to evaluate the distribution properties of the differences between expected and observed data. The pharmacokinetics data were determined according to a two-compartment open model. The following pharmacokinetics parameters were considered: Cl (total body clearance), Vd (volume of distribution), C_{max} (peak drug concentration), T_{max} (time to maximum concentration) and AUC (area under the concentration–time curve)

Statistical methods. The comparison of the pharmacokinetic parameters between the two patient groups (OEM-TACE and FOLFOX) was performed both on the raw experimental data and on standardized data to balance the different doses of oxaliplatin administered between the two groups.

The standardization of data was performed applying the following formula: Z = (principal value – mean value) / standard deviation. The non-parametric Mann-Whitney test and parametric Student's *t*-test were both used to compare both raw experimental data and standardized pharmacokinetic parameters from the two patient groups. Probability values of less than 0.05 were considered significant. Comparison between the ratios of intralesional and extralesional oxaliplatin concentrations in the OEM TACE and FOLFOX groups was performed by nonparametric Mann-Whitney *U*-test. Spearman correlation was applied to test a possible relationship between the time of biopsy (logarithmic scale) and the ratio of concentrations. OS was defined as the interval from the diagnosis to the time of death or the last follow-up. Survival rates were estimated with the use of the Kaplan–Meier method (15) with GraphPad Prism version 5.00 for Windows (GraphPad Software, San Diego, CA, USA).

Toxicity. The patients were evaluated for early and late complications related to the interventional procedure and to the chemotherapeutic agents. Physical examination, blood samples and ultrasound scans were performed after the treatments to identify complications. Any complications were evaluated according to WHO criteria (16).

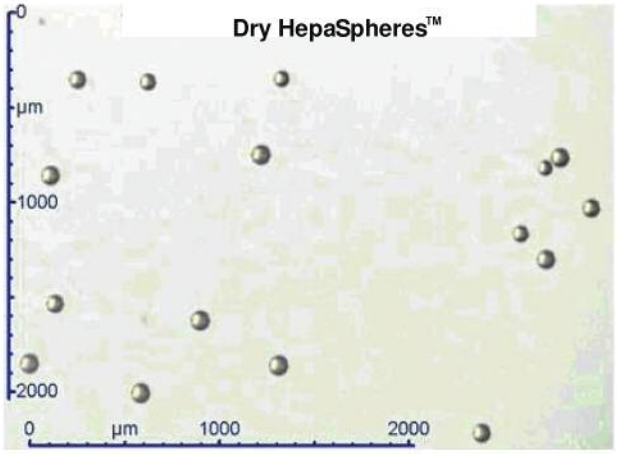
Results

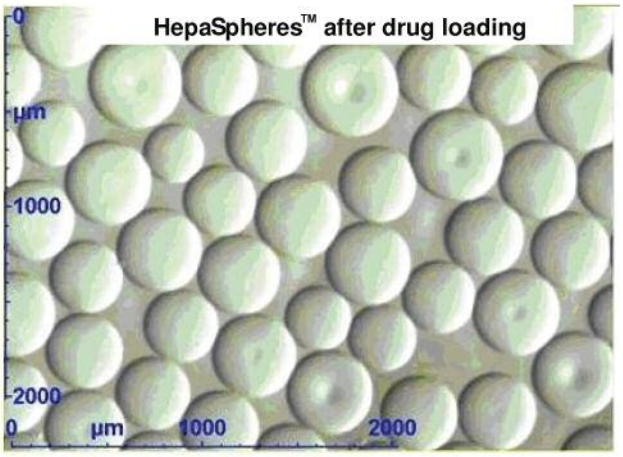
Pre-clinical study. At every time-point (5, 15, 30, 45 min), the platinum concentration in the filtered solution was below the limit of detection (LOD). The data suggested that the microspheres bind oxaliplatin completely. After loading oxaliplatin, the microspheres increased their diameter by approximately four times (Figure 1).

Feasibility, safety and efficacy. The mean follow-up was 34 months (range 6-92 months). The mean time between the OEM-TACE and RFTA was 9.2 days (range 7-14 days). RFTA was not performed in four patients due to high risk of complications (lesions too near to the gallbladder in two patients, too many lesions in the other two). The mean dose of oxaliplatin administrated was 47.7 mg (range 25-100 mg). Overall, the OEM-TACE procedure was well tolerated without serious complications. No OEM-TACE patient exhibited grade 4 complications. The most common adverse event was post-TACE syndrome consisting of low-grade fever, nausea and abdominal pain, occurring in approximately 53.2% of patients and requiring the administration of codeine and acetaminophen for control of right upper quadrant pain and fever. These effects resolved within a few days of the procedure. Two patients with increasingly persistent pain over one week were diagnosed as having acute chemical cholecystitis based on clinical and ultrasound findings. Both were managed with intravenous fluids and pain control drugs. One patient had a mild orticarioid rash after TACE that quickly disappeared after intravenous administration of antihistamine. No hepatic abscess, biliary or vascular damage, or deterioration of hepatic function were documented. No alopecia, bone marrow toxicity or peripheral neurological symptoms occurred. Serum aminotransferases, amylase and lipase were slightly increased soon after the procedure. Only one patient experienced mild pancreatitis with a persistent high increase of pancreatic enzymes and abdominal pain which resolved with conservative therapy. The assessment of response at one month after the last procedure, according to RECIST (14) criteria is shown in Table I. Causes of death were progressive disease in five patients (67%), and intercurrent disease in two patients (33%). The median OS was 40 months (Figure 2). Stable disease was observed in 8 out of the 15 patients (53.3%), a partial response in 2 (13.3%) and intrahepatic or extrahepatic tumor progression in 5 of 15 patients (33.3%). Four patients (one partial response and three stable disease) underwent a ^{18}F-fluorodeoxyglucose PET scan that found no uptake of the tracer, meaning no vital cells in the lesions. In one of these patients, a large intrahepatic cholangiocarcinoma was surgically removed after three cycles of OEM-TACE and complete necrosis was histologically documented.

Pharmacokinetic profile and intratumoral concentration. The pharmacokinetic parameters, as estimated with the P-Pharm software (two-compartment model), are represented in Table II.

The mean C_{max} was significantly higher in the FOLFOX group (1,528 ng/ml ±519) than in the TACE group (632 ng/ml ±280) ($p<0.001$) and it was reached earlier in the TACE group (mean T_{max} 1.53±0.64 h) than in the FOLFOX group (mean T_{max} 3.0±0.58 h) ($p=0.012$). The mean total systemic exposure calculated as the area under the concentration–time curve from 0 to infinite time ($AUC_{0\rightarrow\infty}$) was significantly





		50-106 µm
Dry	mean (µm)	93.91
	range (µm)	64-122
With 5 mg/ml oxaliplatin in Visipaque	mean (µm)	399.86
	range (µm)	260-500
	swelling	4.26

Figure 1. *HepaSpheres™ before (left) and after (right) mixing with Visipaque and oxaliplatin. Lower panel, diameters of HepaSpheres before and after loading.*

higher in the FOLFOX group (2,693 ng/ml min ± 127) than in the TACE group (1,483 ng/ml min ±838) (p=0.028). The statistical data are shown in Table III. The core needle biopsies showed that the ratio between the intra- and extratumoral concentration of the drug was significantly different between the OEM-TACE and FOLFOX groups. The median ratio was 18.53 (1.27-71.2) in the patients treated by OEM-TACE and 1.10 (1.08-1.38) in patients treated by conventional systemic chemotherapy (p=0.014) (Table IV).

The time between oxaliplatin administration and the determination of tissue drug concentration by biopsy did not influence the ratio between the intra- and extratumoral concentration of the drug (r of Spearman=0.24, p=0.48).

Discussion

Only a few published articles have addressed the use of DEM in the treatment of human tumors. DC Bead™ loaded with either doxorubicin or irinotecan has been investigated recently in the treatment of patients with HCC and hepatic colorectal metastasis, respectively. In a study by Varela *et al*. (17) patients with unresectable large or multifocal HCCs were treated with doxorubicin loaded drug eluting beads (DEB-TACE) or conventional TACE (doxorubicin, lipiodol and gelfoam). The results in the patients treated with DEB-TACE were promising, with low side-effects, a high response rate and an advantageous

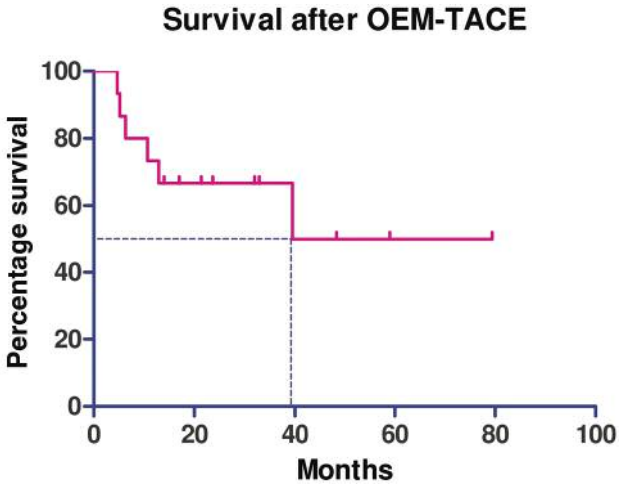


Figure 2. *Kaplan-Meier survival curve.*

pharmacokinetic profile compared with conventional TACE. These results confirmed previous experimental findings *in vitro* and in animal studies (5, 11, 18, 19). In a study by Aliberti *et al*. (20) patients with liver metastases from CRC were treated with irinotecan-eluting bead chemoembolization. The authors reported a decrease of greater than 50% of CEA levels and a significant reduction of contrast enhancement of hepatic

Table II. *Pharmacokinetics parameters in TACE (A) and FOLFOX (B) groups.*

A. TACE group

Dose (mg/ml)	Cl (l/h)	Vd (l)	T_{max} (h)	C_{max} (ng/ml)	AUC (ng/ml min)
30	0.28	121.1	2	360	1785
45	0.18	48.8	3	910	4237
50	0.59	129.9	2	560	1392
45	0.73	119.1	1	650	1030
50	1.1	106.6	1	1170	782
35	0.49	137.9	1	550	1173
50	0.55	117.5	2	620	1502
61	0.61	174.7	1	620	1655
36	0.31	32.1	2	1100	1923
40	0.67	197.7	1	430	985
50	0.66	328.1	1	380	1258
50	0.79	144.6	1	600	1042
28	0.63	237.2	1	240	738
50	0.53	196.5	2	360	1563
50	0.7	80.8	2	930	1190
Mean	0.61	144.9	1.53	632	1483
Median	0.6	129.9	1	600	1258
SD	0.2	74.7	0.64	280	838
CV%	38	51.6	41.7	44	56.6
Max	1.1	328.1	3	1170	4237
Min	0.18	32.1	1	240	739

B. FOLFOX group

Dose (mg/ml)	Cl (l/h)	Vd (l)	T_{max} (h)	C_{max} (ng/ml)	AUC (ng/ml min)
150	0.98	93.2	3.0	1633	2551
160	0.99	98.2	3.0	964	2694
170	1.01	92.9	2.0	1987	2805
Mean	0.99	94.8	3.0	1528	2693
Median	0.99	93.2	2.7	1633	2683
SD	0.02	3	0.58	519	127
CV%	1.54	3.1	21.7	34	5
Max	1.01	98.2	3.0	1987	2805
Min	0.98	92.9	2.0	964	2551

Cl: total body clearance, Vd: volume of distribution, T_{max}: time to maximum concentration, C_{max}: peak drug concentration, AUC: area under the concentration–time curve, CV% coefficient of variation %.

Table III. *Statistical comparison of the pharmacokinetic parameters of the raw (A) and standardized (B) experimental data.*

A. Raw experimental data.

	Group	Mean±SD	Median (Min-Max)	Student's *t*-test (*p-value*)	Mann-Whitney (*p-value*)
Cl	TACE	0.59±0.22	0.61 (0.18-1.10)	0.008	0.021
	FOLFOX	0.99±0.15	0.99 (0.98-1.01)		
Vd	TACE	144.8±74.7	129.9 (32.1-328.1)	0.27	0.11
	FOLFOX	94.77±2.98	93.2 (92.9-98.2)		
T_{max}	TACE	1.53±0.64	1 (1-3)	0.012	0.025
	FOLFOX	2.67±0.58	3 (2-3)		
C_{max}	TACE	632±279	600 (240-1170)	<0.001	0.015
AUC	FOLFOX	1528±519	1633 (964-1987)		
	TACE	1483.67±838.39	1258 (738-4237)		

B. Standardized experimental data.

	Group	Mean±SD	Median (Min-Max)	Student's *t*-test (*p-value*)	Mann-Whitney (*p-value*)
Z_Cl	TACE	–0.009±1.02	0.09 (–1.9 - 2.3)	0.78	0.91
	FOLFOX	0.167±0.76	0.00 (–0.5 - 1)		
Z_Vd	TACE	0.001±1	–0.199 (–1.5 - 2.5)	0.98	0.68
	FOLFOX	–0.011±0.99	–0.53 (–0.6 - 1.1)		
Z_T_{max}	TACE	–0.47±0.64	–1 (–1 - 1)	0.74	0.64
	FOLFOX	–0.33±0.58	0.00 (–1 - 0)		
Z_C_{max}	TACE	0.000±0.99	–0.114 (–1.4 - 1.9)	1	0.95
Z_AUC	FOLFOX	0.000±0.99	0.202 (–1.1 - 0.9)		
	TACE	0.000±1	–0.27 (– 9 - 3.3)		

metastases on CT scan in all the patients. The only reported work with HepaSpheres™ was by Osuga *et al*. (21) who treated six HCC patients with microspheres without chemotherapeutic agents obtaining extensive necrosis of large tumors. Taken together these preliminary reports seem to show some advantages in treating patients with HCCs or hepatic metastases with DEM-TACE instead of conventional TACE. The present study demonstrated that HepaSpheres™ could load up to 400 mg of oxaliplatin and, more importantly, they delivered more into the tumor tissue than into the normal hepatic parenchyma, as well demonstrated by the differences in the drug concentration in the multiple biopsies performed.

Table IV. *Intratumoral and extratumoral concentrations of oxaliplatin.*

Patients	Primary tumor	Intratumoral (μg/g)	Extratumoral (μg/g)	Dose administered (mg)	Ratio	Days between TACE and needle biopsy
1	CRC	315.00	17.00	25	18.50	35
2	Cholangio	72.00	3.80	45	18.90	27
3	Cholangio	17.80	0.25	50	71.20	12
7	CRC	9.60	2.60	100	3.70	11
8	CRC	44.0	2.0	50	22.00	1
9	CRC	1.59	0.51	28	3.11	49
10	CRC	1.38	1.09	50	1.26	18
Control 1	CRC	1.09	0.98	135	1.11	3
Control 2	CRC	1.18	0.90	140	1.31	5
Control 3	CRC	1.03	0.95	125	1.08	10
Control 4	CRC	1.12	1.02	135	1.09	7

The ratios between the intra- and extratumoral concentration of the drug were not influenced by the time at which the biopsies were performed, proof that high concentrations of oxaliplatin persisted a long time in the tumoral tissue. The present pharmacological data were not as attractive as those of the Varela *et al*. (17). The AUC and C_{max} were significantly higher in the FOLFOX than in the TACE group, but only because higher doses of the drug were used in the former group. When the data were standardized in order to balance the difference between drug doses administered in the two different groups, no meaningful statistical difference was found between them (Table IIIB). The divergence between the present pharmacological data and those of Varela *et al*.'s study has many sources, the first being the different type of tumor treated and drug used. At the time of analysis (26 April 2008), six patients had died and eight were still alive. No patient had a complete response: unfortunately RECIST criteria underestimate the rate of objective responses in patients treated with ablative locoregional therapies, as well demonstrated in hepatocellular carcinoma, where responses are better evaluated with European Association for the Study of the Liver (EASL) (22) guidelines. Since the evaluation of tumor necrosis, induced by TACE or RFTA, in colorectal hepatic metastasis and cholangiocarcinoma is not as well determined by contrast imaging as in HCC, EASL guidelines could not be applied to the present patients.

The patient population was not homogeneous and included two different types of cancer, so it was not possible to compare the survival with other traditional treatments. Moreover, some patients were treated with both TACE and RFTA so it was not easy to determine the true role of each procedure in the obtained results. However, the good survival data indicate that treatment with OEM-TACE alone or combined with RFTA deserves to be further investigated.

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