Clinical Value of Immunoscintigraphy in Colorectal Carcinoma Patients: A Prospective Study

Angelika Bischof-Delaloye, Bernard Delaloye, Franz Buchegger, Willy Gilgien, A. Studer, S. Curchod, Jean-Claude Givel, François Mosimann, Jacques Pettavel, and Jean-Pierre Mach

Division of Nuclear Medicine, CHUV, CH-1011 Lausanne; Biochemistry Institute, Lausanne University, CH-1066 Epalinges; Department of Surgery, CHUV, CH-1011 Lausanne, Switzerland

Fifty-seven patients with suspected CEA-producing tumors were studied prospectively by radioimmunoscintigraphy (RIS) using a ^{123}I-labeled anti-CEA monoclonal antibody (MAb) (essentially the F(ab')$_2$ or Fab fragments) and emission computed tomography (ECT). Results of RIS were compared to those of a comprehensive diagnostic study. Final diagnosis was based on surgery, biopsy and autopsy (n = 39) or follow-up findings (n = 18). Three groups of patients were defined: Group A with suspected primary tumors (n = 11), Group B with probable (n = 19) and Group C with questionable (n = 27) tumor relapse. Eighty-eight per cent, 93% and 71% of the anatomic regions studied were correctly identified as being involved, and 97%, 97%, and 87% as being free from tumor in Groups A, B, and C, respectively. In the 27 patients from Group C with no definite diagnosis of relapse, and in whom diagnosis was most difficult, 38 tumor sites were involved. Of these, 21 were detected by both prospective RIS and repeated comprehensive study, six by RIS only and seven by conventional methods only. Four sites remained undetected by both approaches. Ten of the 21 lesions were detected by RIS more than 1 mo earlier than by any other method. Among the seven tumor sites detected by other diagnostic modalities only, three were identified at the time of RIS and four became positive more than 6 mo later. Overall diagnosis was entirely correct in 30, partially correct in 16 and incorrect in six patients studied. RIS with ECT and ^{123}I-labeled anti-CEA MAb allows early detection of recurrence or metastasis of colorectal cancer. It thus contributes to reduced delay between diagnosis and treatment.

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Radioimmunoscintigraphy (RIS) with monoclonal antibodies (MAbs) directed against various tumor associated antigens is of interest in the visualization of primary and secondary tumors. However, most of the studies published are retrospective (*1–10*) even though tumor deposits have been evidenced by RIS only and not by other imaging procedures. Few prospective data have been reported so far on colorectal (*11,12*) and ovarian (*13,14*) carcinomas.

In a previous retrospective study (*9*) we showed that emission computed tomography (ECT) using F (ab')$_2$ and Fab fragments of anti-CEA MAbs, labeled with iodine-123 (^{123}I) made it possible to detect an average of 86% of primary and secondary colorectal tumor sites much more accurately than when ^{131}I-labeled intact antibodies were used (*2, 3, 8*). This work, however, was performed on a selected population of patients. We thus wished to confirm our results in a prospective study and to evaluate the potential role of RIS in the clinical management of colorectal carcinoma patients in comparison with conventional diagnostic procedures.

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For reprints contact: Angelika Bischof-Delaloye, Div. of Nuclear Medicine, CHUV, 1011-Lausanne, Switzerland.

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PATIENTS AND METHODS

Fifty-seven consecutive patients, 39 men and 18 women, aged 36–81 yr (mean 69 yr), with suspected primary or recur-

TABLE 1
Group A: Patients with Suspected Primary With or Without Metastases

Patient no.	Age (yr)	Sex	Primary	Site	Dukes	metas	CEA ng/ml	MAb fragment		Dose mCi
1*	68	F	adenoCA	asc col	C	Liver	64	25/35	F(ab')$_2$	9.4
2	51	F	no known tumor				18	35	Fab	12.2
20*	75	M	adenoCA	asc col	C	None	3	25/35	F(ab')$_2$	5.1
22*	61	F	adenoCA	rectum	D	Liver	205	25/35	F(ab')$_2$	7.2
28*	63	M	epid CA	lung		None	2000	35	Fab	
36	46	M	no known tumor				12	35	Fab	9.9
38*	69	M	adenoCA	rectum	C	Liver, lung	2945	35	Fab	7.5
43*	78	F	adenoCA	caecum	B	None	2	25/35	F(ab')$_2$	5.5
49*	81	F	adenoCA	sigmoid	A	None	1	35	Fab	8.5
51*	64	M	adenoCA	unknown		Bone	262	25	F(ab')$_2$	7.3
52*	70	M	adenoCA	sigmoid	C	Liver	0	25	F(ab')$_2$	8.0
54*	79	M	adenoCA	rectum	B	Liver	7	25/35	F(ab')$_2$	6.7

* Patients with findings confirmed by surgery, biopsy, or autopsy.

rent CEA-producing tumors, underwent RIS after having been informed by the attending physician of the aim and design of our study and having orally consented to participate. The study protocol had been previously accepted by the Ethics Committee of the local Medical School. RIS was performed without knowledge of the clinical presentation and of the probability of disease. After completion of the study, patients were assigned according to clinical criteria to one of the following groups:

Group A. Twelve patients with suspected primary tumors with or without metastases (Table 1).

Group B. Eighteen patients who presented a high probability of recurrence and/or metastases on the basis of symptoms and/or other diagnostic procedures (Table 2). The time from primary tumor resection ranged from 0.2 to 6 yr (mean 1.8 yr).

Group C. Twenty-seven patients with suspected recurrence and/or metastases who presented either with an isolated rise in serum CEA, equivocal results of other diagnostic procedures or with a completely normal diagnostic picture in spite of symptoms (Table 3). The time from primary surgery in this group ranged from 0.1 to 14 yr (mean 2.3 yr).

Details concerning patients, tumor status, serum CEA, MAbs and injected activity are listed in Tables 1 to 3. Three patients were investigated twice: Patient 7 had 2 RIS at a 1-yr interval because of rising serum CEA levels but without clinical or radiologic evidence of tumor. Patient 17 underwent surgery for liver metastasis and was re-studied after 1 yr.

TABLE 2
Group B: Patients with Probable Recurrence and/or Metastases

Patient no.	Age (yr)	Sex	Primary	Site	Dukes	Time from surgery I	CEA ng/ml	MAb fragment		Dose mCi
4	58	M	adenoCA	sigmoid	C	4 mo	28	25/35	F(ab')$_2$	10.7
5*	70	M	adenoCA	sigmoid	C	3 yr	18	25/35	F(ab')$_2$	9.9
10*	58	M	adenoCA	sigmoid	C	2 yr	6	25	F(ab')$_2$	6.2
13*	51	F	adenoCA	sigmoid	A	1 yr	20	35	Fab	5.7
19*	77	F	adenoCA	rectum	C	1 yr	8	25	F(ab')$_2$	3.9
21*†	51	F	adenoCA	sigmoid	C	1 yr	860	35	Fab	4.4
24	73	M	adenoCA	rectum	A	5 yr	16	35	Fab	8.0
25	42	M	adenoCA	lung		1 yr	86	35	Fab	3.8
26*	57	M	adenoCA	sigmoid	B	4 mo	500	35	Fab	5.4
27*	68	M	adenoCA	desc col			1000	35	Fab	4.8
31*	77	M	ad. vill	sigmoid	A	2 yr	230	35	Fab	9.2
33	50	F	adenoCA	desc col	C	5 mo	85	35	Fab	7.7
37‡	63	M	adenoCA	rectum	C	2 yr	36	25/35	F(ab')$_2$	8.0
40	75	F	adenoCA	rectum		6 yr	437	35	Fab	10.3
45*	74	M	adenoCA	sigmoid	C	2 mo	9	35	Fab	8.0
48*	55	F	adenoCA	rectum	A	3 yr	10	25/35	F(ab')$_2$	8.5
57*	54	M	adenoCA	rectum	B	6 mo	4	35	Fab	9.2
61*	64	M	adenoCA	sigmoid	C	1 yr	2	35	Fab	11.5

* Patients with findings confirmed by surgery, biopsy, or autopsy.
† Breast cancer as well as colon cancer.
‡ Second study in the same patient.

TABLE 3
Group C: Patients with no Definite Diagnosis of Tumor Recurrence

Patient no.	Age (yr)	Sex	Primary	Site	Dukes	Time from surgery I	CEA ng/ml	MAb fragment		Dose mCi
3*	79	F	adenoCA	sigmoid	B	3 yr	11	35	Fab	4.8
6*	59	F	adenoCA	transvs	B	1 yr	1	35	Fab	10.1
7a	53	M	adenoCA	caecum	C	1 yr	71	35	Fab	5.6
7b*	54	M	adenoCA	caecum	C	2 yr	110	35	Fab	10.4
8*	69	M	adenoCA	rectum	B	1 yr	37	35	Fab	10.4
9*	63	M	adenoCA	rectum	C	2 yr	125	25/35	F(ab')$_2$	11.4
11†*	63	F	adenoCA	colon		14 yr	194	35	Fab	
12*	70	M	adenoCA	transvs	C	3 yr	174	35	Fab	6.3
14	51	F	adenoCA	sigmoid	C	1 yr	1	35	Fab	
15*	67	M	adenoCA	asc col	C	4 yr	27	35	Fab	11.5
16*	51	M	adenoCA	sigmoid	B	3 yr	82	35	Fab	6.2
17‡*	36	M	adenoCA	desc col	C	1 yr	5	35	Fab	7.1
17§	37	M	adenoCA	desc col	C	2 yr	4	35	Fab	9.0
23	59	F	adenoCA	rectum	C	2 yr	7	25	F(ab')$_2$	5.8
29*	69	F	adenoCA	sigmoid	B	1 yr	28	35	Fab	5.8
30*	51	M	adenoCA	rectum	C	1 yr	46	25	F(ab')$_2$	3.6
32	39	M	adenoCA	sigmoid	C	1 mo	60	35	Fab	10.3
34*	59	M	adenoCA	rectum	B	1 yr	30	25/35	F(ab')$_2$	12.9
35‡	59	M	adenoCA	sigmoid	B	3 yr	23	35	Fab	9.2
37‡	62	M	adenoCA	rectum	C	1 yr	9	25	intact	6.8
39*	47	M	adenoCA	sigmoid	C	7 mo	46	35	Fab	7.9
44*	78	F	adenoCA	transvs	B	1 yr	35	35	Fab	4.1
46	74	M	adenoCA	rectum		3 yr	5	35	Fab	5.0
47	55	M	adenoCA	rectum	C	3 yr	13	25/35	F(ab')$_2$	9.0
55	39	M	adenoCA	asc col	B	1 yr	1	35	Fab	14.6
56	65	M	adenoCA	rectum	A	5 yr	10	35	Fab	9.8
58*	63	M	adenoCA	rectum	D	1 yr	4	35	Fab	9.0

* Patients with findings confirmed by surgery, biopsy, or autopsy.
† Breast and lung cancer as well as colon cancer.
‡§ First and second study in the same patient.

Patient 37 was switched from group C to group B when he was re-studied 8 mo later.

Patients were premedicated per os with potassium iodide (2 × 100 mg per day) 2 days before and 3 days after injection of the iodinated MAbs in order to keep thyroid uptake of free iodine as low as possible (<1% of the injected dose). Gastric uptake and secretion of free iodine were prevented by administration of potassium perchlorate (2 × 400 mg per day) orally for 3 days after injection. In order to minimize the risk of allergic reactions, patients received an anti-histaminic drug (Clemastine, 2 mg, per os) 16 and 1 hr before injection. No skin tests were performed. Patients were kept fasting for at least 4 hr preceding RIS.

All patients received iodine-123- (^{123}I) labeled MAbs. Thirty-eight patients were studied with the Fab fragments of MAb 35, which had given the best results in our retrospective study (*9*). One patient received intact MAb 25, and six received the F(ab')$_2$ fragment of MAb 25 (=B7-25) a new antibody of high affinity for another CEA-specific epitope, distinct from that of MAb 35 (*15,16*). Affinity of MAb 35 is 0.6×10^{10} L/M, that of MAb 25 1.9×10^{10} L/M (*17*). Twelve patients received a mixture of F(ab')$_2$ fragments from MAbs 25 and 35. 0.5 to 2 mg protein were labeled with 3.6 – 14.6 mCi or 135–540 MBq ^{123}I.

Iodine-123 was produced from the ^{127}I (p,5n) xenon-123 (^{123}Xe) reaction and provided by the Swiss Federal Institute for Reactor Research (now the Paul-Scherrer Institute), Würenlingen, Switzerland. The fragments were prepared by pepsin or papain digestion (*15*). Labeling was performed at 4°C by the Iodogen method (Pierce Chemicals, Rockford, IL). Labeled antibodies were separated from free iodine by chromatography on a Sephadex G-25 column (Pharmacia, Uppsala, Sweden) equilibrated in pyrogen-free 0.15*M* saline and sterilized by filtration through a 0.22-μm Millipore filter (Millipore, Bedford, MA). Labeling efficiency ranged from 61% to 85%. Tests for sterility and absence of pyrogenicity were performed in rabbits. All antibody preparations were tested for immunoreactivity by incubation with CEA coupled to cyanogen bromide (CnBr)-activated Sepharose (Pharmacia) as previously described (*15*). The percentage of specific binding of labeled MAbs to insolubilized CEA was 56.03 + 14.56 (mean ± 1s.d.). The results obtained with Fab were lower (46.07 ± 9.97) than with F(ab')$_2$ (62.84 ± 14.39).

After labeling, the MAb was diluted in 100 ml of isotonic saline and infused i.v. over 30 min. At 4–6 hr as well as 20–24 hr after injection, whole-body scans and 2 ECT (one including pelvis and lower abdomen, the other liver and thorax) were obtained in all patients but one, using a dual-head rotating camera (Rota Camera, Siemens, Erlangen, FRG). Some patients were re-studied at 48 hr. Areas of increased uptake were considered positive when their activity rose in comparison with circulating activity between the two

TABLE 4
Results in Numbers of Anatomic Regions* Studied in Each Patient Group

	n	TP	FN	TN	FP
Group A	59	16	2	40	1
Group B	90	26	2	60	2
Group C	130	27	11	80	12
Total	279	69	15	180	15

* Pelvis, abdomen, liver, thorax, bone.
Group A: Patients with suspected primary.
Group B: Patients with probable recurrence.
Group C: Patients with no definite diagnosis of recurrence.
TP: true positives; FN: false negatives.
TN: true negatives; FP: false positives.

recordings and when they could be visualized in at least two different planes (transverse, coronal, sagittal). A total of at least 3×10^6 counts at 4 to 6 hr and 2×10^6 counts at 20 to 24 hr postinjection was stored per region. For both acquisitions about 40–60 min were needed on the first day and 90–120 min on the second day.

Written recordings of RIS were used for comparison with other diagnostic tests (surgery, autopsy or follow-up). Five anatomic regions—pelvis, abdomen, liver, thorax and skeleton of the trunk—were analyzed. Results of RIS were compared to those obtained by a comprehensive diagnostic workup, including at least the following procedures: computer-assisted tomography (CT) of the pelvis, CT and/or sonography (US) of the liver, and chest x-ray. Several patients also underwent barium enema, colonoscopy, CT of the thorax, bone scan, conventional x-ray of the skeleton and magnetic resonance imaging (MRI) of pelvis and liver. Results of this comparison correlated to findings at biopsy (n = 4), surgery (n = 33), follow-up (n = 18) or autopsy (n = 2).

A total of 279 anatomic sites were finally evaluated: pelvis (n = 57), abdomen (n = 54), liver (n = 56), thorax (n = 56), bone (n = 56). Six regions were excluded from analysis for the following reasons: Patient 44 had no liver/thorax ECT because of excessive fatigue. Patient 11 had bone metastases, probably due to concomitant breast carcinoma, because of good response to treatment with Tamoxifen. Since no biopsy could be obtained, the skeleton of this patient was excluded from our evaluation. In Patients 14, 32, and 51 the results regarding abdomen were equivocal since some bowel uptake did not allow precise analysis of this region.

RESULTS

Infusion of labeled antibody was well tolerated by all patients, no adverse reaction of any kind being observed either during or after antibody administration.

Results for each patient group are presented in Table 4 according to the number of anatomic regions affected.

Patients in group A (Table 1) presented eight colorectal, one lung carcinoma and one adenocarcinoma of unknown origin. Six of these patients presented initially with metastatic disease, which explains their rather high serum CEA levels. In Patients 2 and 36, no malignant tumor was evidenced by complete and repeated diagnostic work up in spite of repeatedly elevated serum CEA levels (18 and 12 ng/ml, respectively). In Patient 36, serum CEA was normal when measured after heat extraction of the serum. Both are still free from tumor more than 2 yr after RIS (which was entirely normal) and are thus considered to be true negatives. All primary tumors were correctly identified as well as 4/5 liver,

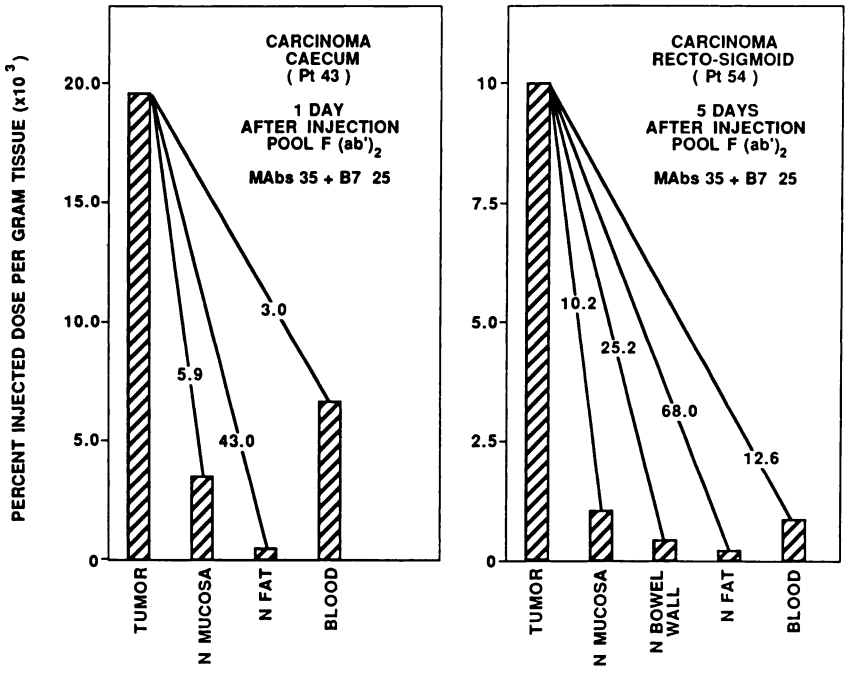

FIGURE 1
Concentration of radioactivity measured in vitro in surgically resected tumor and adjacent normal (N) tissues after injection of F(ab')$_2$ from MAbs 35 and B7-25 labeled with ^{123}I and ^{125}I. Patient 43 (left) was operated from caecum carcinoma 24 hr after injection, Patient 54 (right) from recto-sigmoid carcinoma 5 days after injection. The results are expressed in % of injected dose per g $\times 10^3$ (ordinate). The numbers on the oblique lines indicate tumor to normal tissue ratios. (Normal bowel wall corresponds to bowel wall without mucosa.)

TABLE 5A
Results According to the Number of Anatomic Regions* Studied in Each Patient Group

	Group A	Group B	Group C	Total
Detection rate†	16/18 (89%)	26/28 (93%)	27/38 (71%)	69/84 (82%)
Exclusion rate‡	40/41 (97%)	60/62 (97%)	80/92 (87%)	180/195 (92%)
Accuracy§	51/54 (94%)	91/95 (96%)	107/130 (82%)	249/279 (89%)

* Pelvis, abdomen, liver, thorax, bone.
Group A: Patients with suspected primary.
Group B: Patients with probable recurrence.
Group C: Patients with no definite diagnosis of recurrence.
† True positives with all affected regions.
‡ True negatives over all nonaffected regions.
§ True positives and true negatives over all regions studied.

1/2 lung and 2/2 bone metastases. One patient was erroneously considered to have liver metastases. None of the tumor-free patients showed abnormal accumulation of the labeled MAb.

In two patients operated soon after the injection of a pool of $F(ab')_2$ fragments from MAbs 35 and 25 (B7-25), labeled with ^{123}I and ^{125}I, it was possible to measure radioactivity in the resected tumor and adjacent normal tissues (Fig. 1). In Patient 43, who had a 5 g caecum carcinoma resected 1 day after injection, 0.02% of the injected dose was recovered per gram of tumor, with 5.9 times lower radioactivity concentration in adjacent normal mucosa (Fig. 1, left). In Patient 54, in whom a 5 g recto-sigmoid carcinoma was resected 5 days after injection, the percentage uptake per gram of tumor was 0.01. Tumor activity was ten times higher than that of adjacent normal mucosa (Fig. 1, right).

In group B (Table 2), five cases presented with rather elevated CEA levels, ranging from 230 to 1000. In the other 13 patients, CEA ranged from 2 to 86 ng/ml. RIS correctly identified 9/9 recurrences, 3/4 cases with loco-regional tumor invasion, 9/9 liver, 3/3 lung and 2/3 bone metastases. In one patient a false diagnosis of probable recurrence was made, resulting from uptake in the ascending colon. In the same case, liver metastases were made apparent by RIS and confirmed at laparotomy.

Group C (Table 3) comprises our most difficult cases. Diagnosis of tumor recurrence was not established when RIS was performed. Lower CEA values (1–194) suggest less extensive disease. Seven out of eleven recurrences, 2/4 local extensions, 14/14 liver, but only 4/9 lung metastases were identified by RIS. Four recurrences as well as one lung, one bone, and six liver metastases suggested by RIS could not be confirmed later. According to presently available diagnostic and follow-up findings, these patients were considered to be false-positives.

For obvious reasons, no normal control group could be studied. No conclusions on sensitivity and specificity are, therefore, possible. Our results rather express the ability of this type of radioimmunotomography to detect or to exclude involvement of a given site in patients with primary or secondary colorectal carcinoma.

Table 5A shows detection and exclusion rates as well as accuracy for the total number of sites studied and for each of the patient groups. Table 5B shows only regions which have been verified by surgery, biopsy, or autopsy. These data confirm the results of retrospective studies showing that this method permits sensitive detection of tumor masses when these are large enough to

TABLE 5B
Results According to the Number of Anatomic Regions* Studied and Confirmed by Surgery, Biopsy, or Autopsy in Each Patient Group

	Group A	Group B	Group C	Total
Detection rate†	14/15 (93%)	16/17 (94%)	20/25 (80%)	50/57 (88%)
Exclusion rate‡	12/13 (92%)	15/16 (94%)	21/26 (81%)	48/55 (87%)
Accuracy§	26/28 (93%)	31/33 (94%)	41/51 (80%)	98/112 (88%)

* Pelvis, abdomen, liver, thorax, bone.
Group A: Patients with suspected primary.
Group B: Patients with probable recurrence.
Group C: Patients with no definite diagnosis of recurrence.
† True positives over all affected regions.
‡ True negatives over all nonaffected regions.
§ True positives and true negatives over all regions studied.

TABLE 6
Detailed Results According to the Various Anatomic Regions Studied

	Pelvis	Abdomen	Liver	Thorax	Bone
Detection rate*	21/24 (88%)	9/13 (69%)	27/28 (96%)	8/14 (57%)	4/5 (80%)
Exclusion rate†	29/33 (88%)	39/41 (95%)	21/28 (75%)	41/42 (98%)	50/51 (98%)
Accuracy‡	50/57 (88%)	48/54 (89%)	48/56 (86%)	49/56 (88%)	54/56 (96%)

* True positives over all affected regions.
† True negatives over all nonaffected regions.
‡ True positives and true negatives over all regions studied.

be suspected from symptoms or other diagnostic procedures (groups A and B). In group C patients, in whom signs of possible tumor growth were much more discrete, 82% of 130 anatomic sites studied were correctly identified as being affected by or free from tumor.

Table 6 indicates detection and exclusion rates attained, as well as accuracy according to the anatomical regions studied. The highest incidence of tumor involvement was observed in liver (50%) and pelvis (42%), followed by thorax (25%), abdomen (24%) and finally bones (9%). Best results were obtained for the pelvis, the presence or absence of tumor being ascertained in 21 of 24 patients (88%) and in 29 of 33 patients (88%), respectively. The highest detection rates (96%) were observed for liver metastases, but the false-positive rate was 25%. Lung metastases were the most difficult to detect (57%) by RIS.

We have also analyzed the results for each individual patient and have categorized them as being correctly positive when all tumor involvement sites had been identified, and as correctly negative when no positive uptake had been shown in patients free from tumor. In patients with one or more tumor sites correctly identified, but with additional false-positive and/or false-negative sites, RIS diagnosis was considered as being partially correct. It was defined as being incorrect in patients with false-positive and/or false-negative results only (Table 7).

Considering the total number of patients studied, diagnosis was entirely correct in 63% and partially correct in 25%. The percentage of correct diagnoses was even higher for groups A (75%) and B (83%). In group C, however, diagnosis was entirely correct in less than one-half (44%), partially correct in one-third (33%) of the patients, and incorrect in six patients (22%).

We have analyzed group C patients in more detail. There were 38 positive tumor sites, 14 in the liver, nine in the pelvis, nine in the thorax and six in the abdomen.

Table 8 compares the findings of RIS and comprehensive workup performed in these patients within 1 mo of RIS and subsequently repeated at various time intervals. A total of 21 sites were evidenced by RIS and the conventional approach, four remained undetected by both, six were positive with RIS only and seven were detected by one or more of the other diagnostic procedures but not by RIS.

Time course of lesion detectability was studied with respect to comprehensive diagnostic workup which was repeated, if initially negative or equivocal, at various time intervals. Final results were compared to the findings of RIS which was performed only once. Table 9 shows the time interval during which lesions could be demonstrated by at least one of the conventional methods, compared to time of diagnosis by RIS. Ten of 21 lesions which were evidenced by both could be detected by RIS between 1 and 6 (n = 6) or even more than 6

TABLE 7
Results According to Patient Studies in Each Group

	n	Correct positive*	Correct negative†	Partially correct‡	False§
Group A	12	7	2	3	0
Group B	18	14	1	2	1
Group C	27	10	2	9	6
Total	57	31	5	14	7

* Studies with affected and nonaffected sites correctly identified.
† Tumor free patients correctly identified.
‡ In addition to correctly classified regions presence of false-positive and/or false-negative sites in the same patient.
§ Studies with only false-positive and/or false-negative results.

TABLE 8
Prospective RIS Versus Comprehensive Diagnostic Workup Performed at the Time of RIS and Repeated During Follow-up in Patients with Questionable Recurrence (Group C)

	Comprehensive workup +	Comprehensive workup −
RIS +	21	6
RIS −	7	4

TABLE 9
Time Course of Detection of Lesions by Conventional Methods Within 1 mo of RIS and During Subsequent Follow-up

	Comprehensive diagnostic workup +			
	<1 mo	1–6 mo	>6 mo	All
RIS +	11	6	4	21
RIS –	3	0	4	7

mo (n = 4) earlier than by any of the standard methods. Among the seven lesions that were silent at RIS, only three were detected by other methods at the time of RIS, four being apparent more than 6 mo later. All four were lung metastases. In none of these patients could RIS be repeated after the appearance of lung metastases on CT. The question remains open whether these lesions would have been detected by RIS if performed at a more advanced stage. Among the 16 sites which were detected at RIS only (n = 6) or earlier than with conventional methods (n = 10) there were seven liver involvements, six local recurrences, two lung metastases and one peritoneal extension.

Figure 2 shows one of the cases in which the diagnosis of liver metastases was made before CT. This 63-yr-old patient (Patient 9) had undergone surgery for carcinoma of the rectum (Dukes stage C) 2 yr before RIS, followed by resection of local recurrence 5 mo later. Two months before RIS, liver CT was performed because of rising serum CEA (125 ng/ml) and the result was considered normal (left). Because of RIS evidence of liver metastasis, which appeared as a single hot spot on a transverse section of the liver (center), CT was repeated 3 wk later, and remained normal. The next liver CT, performed 4 mo after RIS (right) demonstrated metastasis, which was subsequently confirmed by surgery. Levels of tomographic sections, however, were not precisely the same, since these were routine CT examinations in which exactly corresponding sections often cannot be obtained for repeat studies.

Figures 3 and 4 illustrate the case of a 62-yr-old patient (Patient 37[‡§]) who underwent surgery 1 year before the first RIS for carcinoma of the rectum (Dukes stage C). Diagnostic workup, performed because of slightly rising serum CEA (9.3 ng/ml), showed no evidence of tumor recurrence. RIS strongly indicated tumoral involvement of the right retrovesical space (Fig. 3, panel A) and a sub-phrenic liver metastasis (Fig. 4, panel A). Moderate asymmetry shown by pelvic CT (Fig. 3, panel B) was not considered indicative of malignancy. Eight months later serum CEA had risen to 36 ng/ml, and pelvic CT showed a large mass (Fig. 3, panel D) which corresponded to a region of high antibody uptake on RIS (Fig. 3, panel C). The liver metastasis remained visible on RIS (Fig. 4, panel B), but no other diagnostic procedure concerning the liver was performed at that time. Nine months later, a 9-cm subphrenic liver mass was demonstrated ultrasonically.

DISCUSSION

The overall detection rate of radioimmunotomography (82%) appears quite high, particularly in patients (groups A and B) with clinically significant disease (89% and 93%, respectively). It is comparable with that of our retrospective study (*9*). In a smaller series of patients with suspected recurrent ovarian carcinoma and investigated by RIS, a slightly lower percentage (72%) has been reported by Chatal et al. (1987). In our series of patients in whom diagnosis of relapse could not definitely be established prior to RIS (group C), 71% of the affected and 87% of the disease-free anatomic regions have been correctly identified.

Regarding anatomic regions, the highest tumor inci-

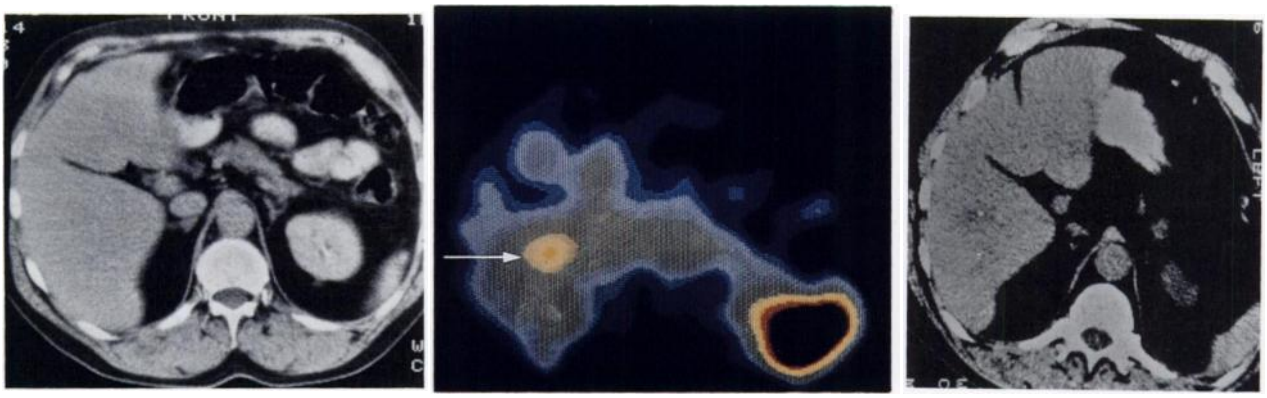

FIGURE 2
Liver CT scans and radioimmunotomography of a patient having undergone surgery for rectal carcinoma 2 yr before RIS and for local recurrence 5 mo later. CT performed 2 mo before RIS (left) was normal. Liver metastasis (arrow) shown on ECT with ^{123}I-labeled anti-CEA antibodies (center) remained undetected by CT 3 wk later; it became visible only at the following control, performed 4 mo after RIS (right).

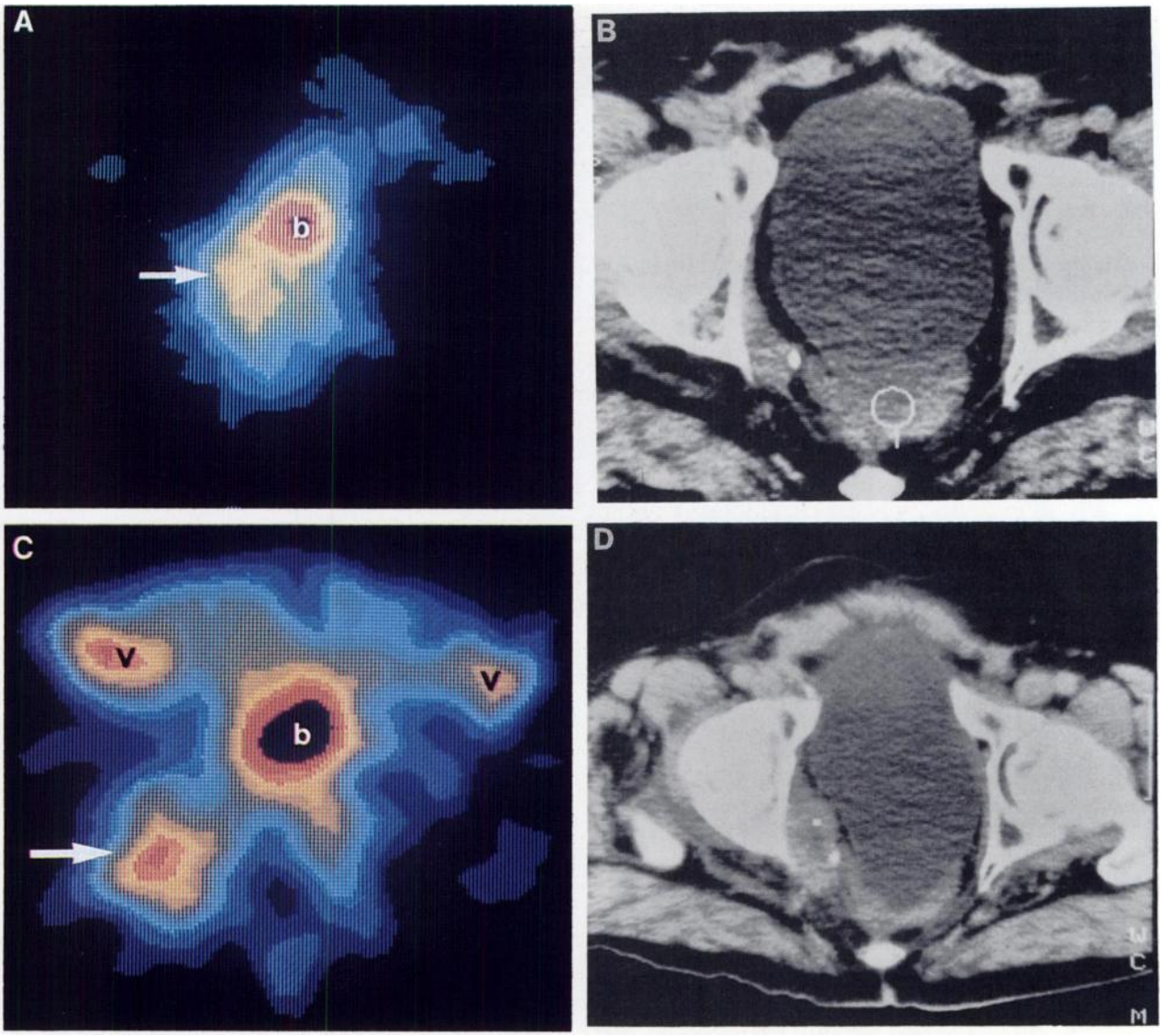

FIGURE 3
ECT of the pelvis obtained 48 hr after injection of ^{123}I MAbs 35 Fab (panel A) shows accumulation in the right retro-vesical space (arrow), whereas moderate asymmetry on CT (panel B) was not considered indicative of malignancy 1 yr after primary surgery for rectal carcinoma. A repeat study, performed 8 mo later, clearly showed the tumor (arrow) behind the bladder (b) on ECT (panel C) as well as on CT (panel D). More intense circulating activity in the iliac vessels (v) on the second RIS (panel C) is due to the shorter interval from injection (24 hr).

dence was found in liver (50%) and pelvis (42%) as expected. CT is considered to be accurate in detecting recurrence of local colorectal carcinomas (*18,19*), but it often does not differentiate tumors from inflammation and fibrosis (*20*). The overall detection rate of local recurrence was 80% in our present series of patients studied by RIS. It thus compares favorably with CT. MRI may be superior to CT in tissue characterization. From a recent study of 29 patients with suspected local recurrences after surgery for rectal carcinoma (*21*), MRI indeed appears to be superior to CT. There is a lack of false-positive or false-negative results with MRI against four false-positive and two false-negative studies with CT in this series. In six patients (21%), however, no clear-cut diagnosis could be made with MRI. Highly accurate RIS should thus improve results obtained by CT and MRI alone, particularly if the latter are guided by RIS. Combined interpretation of functional and anatomic information might increase diagnostic accuracy.

Detection of retro-vesical tumor sites may be difficult when iodine-labeled MAbs are used due to the usually high radioactivity of the bladder. This is particularly true when bladder configuration is changed after colorectal surgery, adding to difficulties of interpretation. New techniques make it possible to obtain iodine-labeled MAbs which are less prone to dehalogenation and may resolve this problem. They are presently under

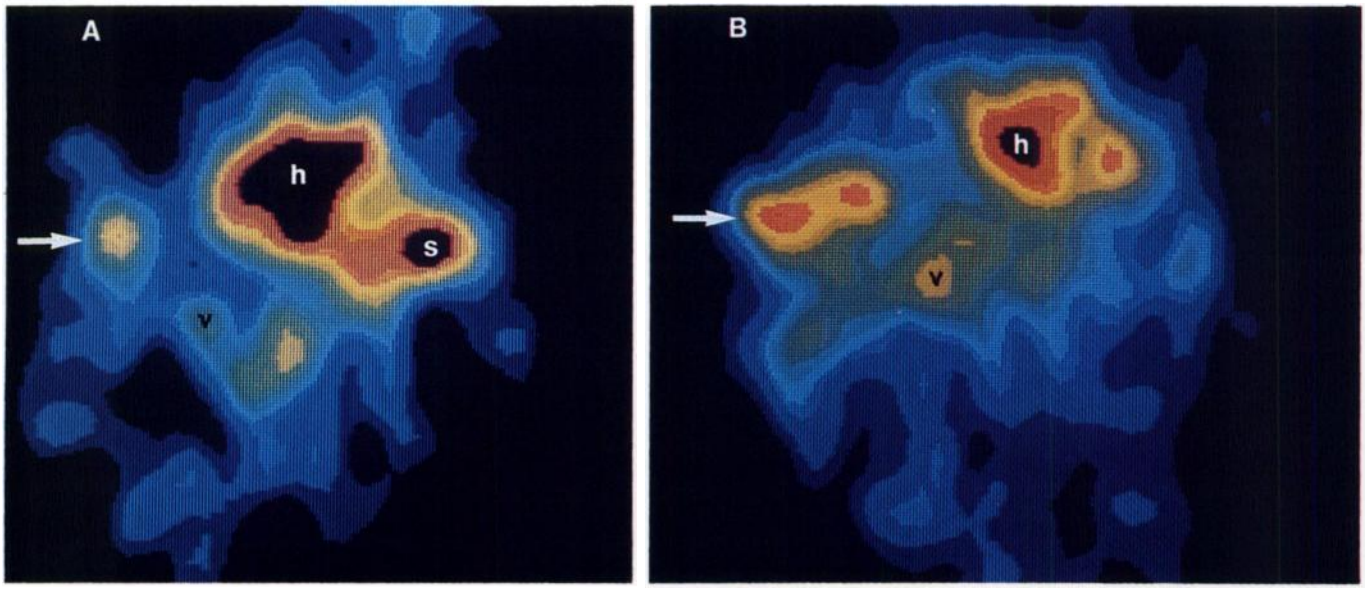

FIGURE 4
In the same patient as in Figure 3 coronal sections of the upper abdomen and lower chest showed accumulation of the anti-CEA MAbs in the subphrenic area (arrow). It is discrete on the first (panel A) and more pronounced on the second (panel B) study, which was performed 8 mo later. At the time of the first RIS, US did not show the metastasis; 9 mo after the second study it demonstrated a large subphrenic mass with extensive central necrosis. (h: heart, s: stomach, v: vascular activity.)

investigation in the group of Zalutsky (*22*), Wilbur (personal communication) and in our group (Kurth et al: in preparation).

Use of indium-111- (^{111}In) labeled MAbs for which urinary excretion rates are much lower, is another alternative. The main drawback of presently available ^{111}In-labeled MAbs is the rather high nonspecific uptake by the reticulo-endothelial system, particularly in the liver. The high incidence of liver metastases in our patients raises the problem of detection of liver metastases by RIS. The high liver uptake observed with ^{111}In-labeled MAbs (*23–26*) makes it difficult to detect liver involvement. Concomitant administration of excess amounts of unlabeled antibody decreases nonspecific uptake of ^{111}In-labeled MAbs in spleen and bone marrow (*27*) and even in liver (*28*), particularly when the labeled antibody is administrated as Fab' fragment (*29*).

Technetium-99m- (^{99m}Tc) labeled fragments of anti-CEA (*30*) and anti-melanoma (*31*) MAbs also show lower liver uptake. It remains to be proved to what extent these techniques allow detection of very small metastases. In our own preliminary experience with the same ^{99m}Tc-labeled anti-CEA MAb as used by Baum et al. (*30*) imaging of liver metastases was not as accurate as with the ^{123}I anti-CEA MAbs. In our present series, in group C patients, all 14 cases of liver involvement could be visualized and seven were detected more than 1 mo before they were visible by CT and/or sonography.

For clinical management of such patients it is important to detect liver involvement as early as possible. Indeed, a multi-institutional study on resection of liver metastases of colorectal carcinoma (*32*) shows that, among many other parameters studied, a serum CEA level of less than 5 ng/ml before resection is the best indicator of good prognosis, particularly if the pathologic margin of the resected specimen is larger than 1 cm and the number of metastases is lower than 3. All these factors indicate limited disease. CT and US can recognize metastatic liver involvement with high sensitivity and specificity (*33,34*). They may, however, fail to detect occult liver disease. In a recent comparison of these two imaging modalities with laparotomy findings (*35*), US depicted only 37% and CT only 55% of lesions with a mean diameter of 1 cm or less, while sensitivity was 80.5% by US and 92% by CT for larger lesions. The overall sensitivity was 52.3% for US and 57.1% for CT in that study.

The detection rate (96%) of liver metastases with RIS in our present series and the fact that a significant number of these were detectable earlier with RIS than with any other conventional technique appears promising. This high detection rate, however, was accompanied by a rather elevated false-positive ratio (25%) resulting from difficulties in distinguishing nonspecific liver uptake from small metastases, especially in patients with concomitant liver disease such as fatty degeneration or cirrhosis. Combined with CT, sonography, or MRI, radioimmunotomography may improve early detection of liver metastases in patients with colorectal carcinoma.

In spite of our promising results, a certain number of problems remain, such as immunogenicity and low absolute tumor uptake of MAb. Multiple injections even of Fab fragments of murine MAbs may lead to the production of human anti-mouse antibodies (HAMA) (*36,37*). The use of monoclonal antibodies raised in other species (e.g., porcine anti-CEA MAbs) (*38*) may be a first step to overcome this problem. An alternative approach is to decrease the immunogenicity by using mouse/human chimeric MAbs (*39*). A chimeric form of the murine MAbs 25 used in this study has been prepared (*17*) and has given promising results in patients (*40*). This, however, might not solve the problems of human responses against antibody variable regions which will remain murine in origin. Almost entirely human antibodies prepared by oligonucleotide-directed mutations of hyper-variable regions of human IgG genes, according to the nucleotide sequences of selected murine hybridomas (*41*), might be the ultimate reagent for broad clinical application of RIS.

Another limitation of RIS is the low absolute tumor uptake of labeled MAbs, even if with the MAbs used here, it was in the order of 0.01–0.03 % of the injected dose per gram of tumor (unpublished data), thus somewhat higher than currently reported. This low uptake requires long acquisition times if statistically valid data are to be obtained. This is particularly important when performing ECT which we prefer to planar imaging, since it does not require background subtraction for image enhancement, even if it is cumbersome for patients and staff. Results may also be directly compared to those of other tomographic methods (US, CT, MRI).

Radioimmunotomography using ^{123}I-labeled fragments of anti-CEA MAbs may already improve the management of selected patients having undergone surgery for colorectal carcinoma since it allows detection of early liver involvement and differentiation between fibrosis and inflammation from local recurrence. Further technological developments (*17,39–42*) may improve the quality of surveillance of colon carcinoma patients with RIS in the near future.

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