Improved Radioimmunoimaging of Human Tumor Xenografts by a Mixture of Monoclonal Antibody F(ab')$_2$ Fragments

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F(ab')$_2$ fragments of monoclonal antibodies (MAbs) GA 73-3 and CO 29.11, with specific binding reactivity in vitro to human tumors of the gastrointestinal tract, were radioiodinated and injected into nude mice bearing human colon carcinoma xenografts. Fragments of both MAbs preferentially localized in tumor tissue compared with normal mouse tissue, as determined by differential tissue counting of radioactivity. The fragments localized specifically only in those tumors to which they bind in vitro and not in unrelated tumors. Radiolabeled fragments of an anti-hepatitis virus MAb did not localize in the tumors. Whole-body scintigraphy demonstrated tumor localization with ^{131}I-labeled fragments without background subtraction. Best tumor contrast, as quantitated by analyzing digital computer scans, was obtained between Days 2 and 5 after injection. Tumor contrast was significantly enhanced when a mixture of both MAb F(ab')$_2$ fragments was used. The biologic half-life of the MAb mixture in the tumor was significantly greater than that of either MAb alone, suggesting the use of the MAb mixture in radioimmunotherapy.

J Nucl Med 27:1739–1745, 1986

The feasibility of using radiolabeled monoclonal antibodies (MAbs) in localizing tumors for detection by external scintigraphy is well documented [reviewed in (*1–3*)]. However, localization is occasionally not successful due perhaps to the detection by a single labeled MAb of an insufficient percentage of tumor cells among this presumably heterogenous population and/or to an insufficient number of radiolabeled MAb molecules bound per cell. Mixtures of MAbs directed against different tumor-associated antigens might overcome these difficulties since they would be expected to label a greater percentage of different cells within the tumor and/or increase the density of radiolabeled MAb on an individual tumor cell. In either case, tumor radioimmunodetection would be improved.

One animal study using mixtures of polyclonal intact antibodies has shown potentiation of tumor localization (*4*). A clinical investigation applying combinations of F(ab')$_2$ fragments of MAbs to a limited number of patients has demonstrated a tendency towards increased tumor detection sensitivity (*5*). There has been no controlled comparative study, however, of MAbs used alone and in combination.

In the present study, we have compared the potential of two recently developed MAbs, GA 73-3 (*6*) and CO 29.11 (*7*), which detect gastrointestinal cancer-associated antigens to localize human colorectal cancer (CRC) xenografts in nude mice when administered alone or in combination.

F(ab')$_2$ fragments of the two MAbs were used, since the imaging characteristics are reportedly superior to both intact antibody (*1,8–13*) and Fab fragments (*13*).

MATERIALS AND METHODS

Human Tumor Cell Lines

The CRC cell line SW-948 and the melanoma cell line WM-9 have been described elsewhere (*14,15*); both are maintained in culture in our laboratory.

Mice and Xenografts

Four- to 6-wk-old nude mice (nu/nu BALB/c background) were injected subcutaneously with 1.5×10^7 human tumor cells in the upper dorsal region. All mice received 0.1% (v/v) KI in their drinking water through-

Received Oct. 5, 1985; revision accepted Apr. 23, 1986.

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out the experiment beginning 48 hr before radiolabeled antibody administration in order to block uptake of free radioiodine by the thyroid gland.

Mouse MAbs

Anti-gastric cancer MAb GA 73-3 (IgG2a) binds to a protein antigen with three molecular weight components of 29k, 30k, and 37k daltons on adenocarcinomas of the gastrointestinal tract, prostate, ovary, bladder, lung, and breast (*6*). The antigen is shed by CRC cells into the culture medium but has not been detected in the sera of colorectal cancer patients (Herlyn M, unpublished data). Anti-CRC MAb CO 29.11 (IgG1) binds to sialylated Lewis a antigen, expressed by adenocarcinomas of the colon, stomach, pancreas, and urinary bladder, but to a different epitope and with a higher binding affinity than the MAb CA 19-9; the antigen is shed both in vitro by CRC cells and in vivo into cancer patients' blood (*7*).

Monoclonal anti-hepatitis virus antibody A5C3* (IgG2a) was used as a negative control antibody.

Purification of MAbs and Production of F(ab')$_2$ Fragments

MAbs were purified from ascitic fluid on protein A-Sepharose columns (*16*). F(ab')$_2$ fragments were prepared and purified as described previously (*11*).

Radiolabeling of F(ab')$_2$ Fragments

F(ab')$_2$ fragments were labeled with iodine-131 (^{131}I) or iodine-125 (^{125}I) using the Iodogen method (*17*). Briefly, 25-μl aliquots of Iodogen (1,3,4,6-tetrachloro-3α,6α-diphenylglycoluril)† at 400 μg/ml in chloroform were evaporated to dryness at 37°C in 10 × 75-mm glass tubes. Sodium iodine-131 or sodium iodine-125 (for protein iodination)‡ and 25–40 μg of protein in 100 μl PBS were added to the tubes, and incubated for 1.5 min at room temperature. The radiolabeled protein was then separated from free iodine on Dowex§ columns equilibrated with PBS containing 5% agammaglobulinemic horse serum. For organ distribution, F(ab')$_2$ fragments were labeled to 1.7–6.4 μCi ^{131}I or ^{125}I per μg of protein. For radioimmunoimaging, specific activities of 6.8–14 μCi ^{131}I per μg of protein were obtained.

Antibody Binding Assay

Labeled F(ab')$_2$ fragments of the anti-tumor MAbs and the A5C3 control MAb were tested for their binding specificity in vitro using SW-948 CRC and WM-9 melanoma cells as targets (*12*).

In Vivo Antibody Distribution and Radioimmunoimaging

Labeled F(ab')$_2$ fragments were injected i.p. into nude mice bearing 7-day-old tumors. Intraperitoneal injections are easier to perform than i.v. injections, and in previous experiments we have found no differences in the tumor localization by MAb following either i.p. or i.v. administration (unpublished results).

For biodistribution studies, the mice received 10 μCi (~1.7 μg) of ^{131}I-labeled F(ab')$_2$ fragments. Another group of mice was injected simultaneously with 10 μCi each of ^{131}I-labeled specific and ^{125}I-labeled unrelated (A5C3 anti-hepatitis virus) F(ab')$_2$ fragments. Two to five days later, the mice were killed and dissected. Tumors, blood, visceral organs, and muscle samples were weighed and assayed for radioactivity. The results are expressed as: (a) ratios of specific activity of F(ab')$_2$ fragments in tumor to that in normal mouse tissues [(cpm/g in tumor)/(cpm/g in normal tissue)]; (b) percentage of injected radioactivity per gram of tissue, corrected for physical decay; (c) biologic half-life ($T_{\frac{1}{2}}$biol.) in tumors and tissues; and (d) localization index, i.e., the ratio of specific (^{131}I) to nonspecific (^{125}I) activity in tumor and in tissues divided by the same ratio in the blood (*18*).

For imaging studies, 100–200 μCi (6–26 μg) of ^{131}I-labeled F(ab')$_2$ fragments were injected i.p. into each mouse. Images were obtained daily for up to 6 days after injection. The mice, usually two to six at a time, were anesthetized and images obtained in posterior and lateral views using a large field-of-view scintillation camera¶ equipped with a medium energy parallel-hole collimator and interfaced to a computer.** Each image was digitized in a 128-by-128-pixel matrix and stored on disk for later analysis and display. No background subtraction or computer smoothing was used.

Scintigraphic analysis was performed on posterior view digital computer images. Specific regions of interest (ROIs) were selected to integrate the counts present in tumor, total body, and rest of the body, i.e., counts in total body minus counts in tumor, at several time intervals. ROIs of the same size and shape were applied to each individual tumor and rest of the body, respectively. Integrated counts were normalized to time and unit area to yield counts per minute (cpm) per pixel. To determine $T_{\frac{1}{2}}$biol. in tumor, total body, and rest of the body, values of cpm/pixel were corrected for tracer decay. Best linear fits were applied excluding the values obtained within 24 hr after injection to avoid the effect of the rapid first component of MAb F(ab')$_2$ clearance from the blood and different tissues.

In addition to digital computer scanning, analog images were simultaneously produced on transparent and polaroid films.

Statistics

The statistical significances of differences ($p < 0.05$) was determined using the Student's t-test and the Mann-Whitney U-test.

RESULTS

In Vitro Binding Reactivity of Radiolabeled F(ab')$_2$ Fragments of MAbs GA 73-3 and CO 29.11

Iodinated F(ab')$_2$ fragments of MAb GA 73-3 demonstrated a higher binding reactivity to SW-948 CRC

target cells than did those of MAb CO 29.11. Maximum binding to target cells was significantly higher ($p < 0.002$, Mann Whitney U-test) for labeled GA 73-3 $F(ab')_2$ as compared with labeled CO 29.11 $F(ab')_2$ (35 to 60% and 26 to 42% binding, respectively, in different experiments). The differences in the values obtained for each MAb in different experiments most likely are due to differences in the specific activities of the iodinated preparations and/or differences in the antigen expression by the target cells at different in vitro passages and/or cell cycle stages. Neither of the fragments bound to melanoma cells (<1% maximum binding). Iodinated $F(ab')_2$ fragments of monoclonal anti-hepatitis virus antibody did not bind to SW-948 CRC cells (<2% maximum binding).

In Vivo Distribution of ^{131}I-Labeled $F(ab')_2$ Fragments of MAbs GA 73-3 and CO 29.11

Nude mice bearing SW-948 human CRC tumors were injected i.p. with ^{131}I-labeled $F(ab')_2$ fragments of MAb GA 73-3 or CO 29.11. Tables 1 and 2 list the tumor-to-tissue ratios of radioactivity, specific activities of tissues, and localization indices obtained with MAbs GA 73-3 and CO 29.11, respectively. $F(ab')_2$ fragments of both MAbs demonstrated preferential localization in CRC tumors as reflected by high tumor-to-tissue ratios of radioactivity as well as high specific activities in tumor tissues. Further evidence of specific localization of both $F(ab')_2$ fragments in CRC tumors was established by the localization indices derived from relevant antibody/unrelated antibody (A5C3 anti-hepatitis virus MAb) ratios in tumor tissue relative to blood. These were between 2.17 (Day 2) and 6.29 (Day 5) for GA 73-3 and between 2.28 (Day 2) and 4.32 (Day 4) for CO 29.11, indicating an increased localization of the relevant MAb to the tumor by 5 and 4 days, respectively. By comparison, the localization indices in all normal mouse tissues tested were between 0.33 and 1.31 for MAb GA 73-3 and 0.25 and 0.75 for MAb CO 29.11, reflecting the similar distribution of both tumor-specific and unrelated $F(ab')_2$ fragments in these tissues on all days tested (not shown).

TABLE 1
Biodistribution of ^{131}I-Labeled $F(ab')_2$ Fragments of MAb GA 73-3 in Nude Mice Bearing SW-948 CRC Tumors*

Tissue	Tumor/tissue ratios of radioactivity [(cpm/g tumor)/(cpm/g tissue)]	Percent antibody dose injected/g of tissue	Localization index
Tumor	—	0.45 ± 0.17†	6.29 ± 3.35†
Blood	10.3 ± 4.5†	0.04 ± 0.01	—
Small intestine	53.8 ± 28.9	0.01 ± 0.003	1.00 ± 0.29
Colon	41.7 ± 23.2	0.01 ± 0.003	1.11 ± 0.64
Stomach	19.0 ± 14.1	0.03 ± 0.01	1.31 ± 0.91
Spleen	33.4 ± 18.8	0.01 ± 0.004	0.89 ± 0.64
Kidney	9.5 ± 3.8	0.05 ± 0.004	1.22 ± 0.87
Liver	27.3 ± 13.4	0.02 ± 0.003	1.21 ± 0.52
Lung	17.9 ± 7.6	0.03 ± 0.004	1.10 ± 0.29
Heart	39.7 ± 17.8	0.01 ± 0.005	0.93 ± 0.44
Muscle	99.1 ± 51.1	0.01 ± 0.002	1.17 ± 0.46

*At 5 days following simultaneous injections of 10 μCi (1.7 μg) each of ^{131}I-labeled GA 73-3 $F(ab')_2$ and ^{125}I-labeled A5C3 $F(ab')_2$.
†Mean ± s.d. of five animals.

TABLE 2
Biodistribution of ^{131}I-Labeled $F(ab')_2$ Fragments of MAb CO 29.11 in Nude Mice Bearing SW-948 CRC Tumors*

Tissue	Tumor/tissue ratios of radioactivity [(cpm/g tumor)/(cpm/g tissue)]	Percent antibody dose injected/g of tissue	Localization index
Tumor	—	0.57 ± 0.13†	4.32 ± 2.64†
Blood	5.4 ± 1.3†	0.11 ± 0.007	—
Small intestine	32.6 ± 9.1	0.02 ± 0.003	0.65 ± 0.28
Colon	33.8 ± 15.9	0.02 ± 0.005	0.41 ± 0.09
Stomach	13.5 ± 12.7	0.06 ± 0.03	0.65 ± 0.16
Spleen	11.4 ± 5.8	0.06 ± 0.02	0.38 ± 0.19
Kidney	8.6 ± 2.8	0.07 ± 0.01	0.49 ± 0.16
Liver	12.4 ± 5.1	0.05 ± 0.01	0.39 ± 0.12
Lung	10.7 ± 4.0	0.06 ± 0.02	0.51 ± 0.17
Heart	30.6 ± 9.0	0.02 ± 0.005	0.73 ± 0.10
Muscle	76.3 ± 20.9	0.01 ± 0.002	0.75 ± 0.41

*At 4 days following simultaneous injections of 10 μCi (1.7 μg) each of ^{131}I-labeled CO 29.11 $F(ab')_2$ and ^{125}I-labeled A5C3 $F(ab')_2$.
†Mean ± s.d. of four animals.

The selectivity and specificity of binding of ^{131}I-labeled $F(ab')_2$ fragments of both MAbs to human tumor cells, as demonstrated by differential tissue counting, suggested the feasibility of radioimmunoimaging the targeted tumors.

Tumor Localization of ^{131}I-Labeled $F(ab')_2$ Fragments of MAb GA 73-3 or CO 29.11 by Radioimmunoimaging

Radioimmunoimaging was performed in nude mice grafted with human SW-948 CRC tumors and injected with 100 μCi each of ^{131}I-labeled GA 73-3 $F(ab')_2$ (6–10 μg) or CO 29.11 $F(ab')_2$ (7–8 μg). Digital computer images as well as analog images on transparent and polaroid films of mice bearing SW-948 CRC tumors weighing between 40 and 200 mg showed localization of labeled $F(ab')_2$ fragments of GA 73-3 and CO 29.11 in all of the 16 and 19 animals examined, respectively. Tumor localization was possible without background subtraction. As early as 24–48 hr after $F(ab')_2$ administration, tumors were visible on both lateral and posterior view scintigrams, becoming increasingly clear throughout the study as radioactivity in the rest of the body decreased. Best tumor contrast, as quantitated by analyzing digital computer images, was obtained between Days 2 and 5 after MAb injection. The maximum ratios of cpm/pixel in the tumors to the rest of the body obtained after injection of GA 73-3 $F(ab')_2$ fragments ranged from 1.75 to 3.71 for the various mice tested. Those of CO 29.11 $F(ab')_2$ fragments ranged from 1.50 to 2.80. Transient bladder and stomach activity was

observed in a few animals during the early period (1–2 days) of the radioimaging. The thyroids showed no uptake of radioactivity on any of the days tested.

Two groups of three mice each bearing WM-9 melanoma xenografts similar in size to SW-948 CRC tumors were given 100 μCi each of ^{131}I-labeled F(ab')$_2$ fragments of GA 73-3 or CO 29.11, and imaging was performed as described for CRC tumors. None of the melanomas were visible at any time after injection of the F(ab')$_2$ fragments. The maximum ratios of cpm/pixel within the tumor to rest of the body ranged from 0.9 to 1.3.

In Vitro Binding of Mixtures of Radiolabeled F(ab')$_2$ Fragments of MAbs GA 73-3 and CO 29.11

Adding mixtures of equal amounts of iodinated F(ab')$_2$ fragments of both MAbs to SW-948 CRC target cells in most experiments resulted in an increase in the maximum amount of bound labeled protein as compared with either component alone. The percentage binding in the fragment mixture (58–89% in five different experiments) exceeded the percentages bound of either component (35–43% of GA 73-3; 27–33% of CO 29.11), indicating an additive or synergistic binding effect. These differences were statistically significant ($p < 0.02$, Mann-Whitney U-test). The mixture did not bind to WM-9 melanoma cells.

Tumor Localization of a Mixture of ^{131}I-Labeled F(ab')$_2$ Fragments of MAbs GA 73-3 and CO 29.11 by Radioimmunoimaging

Figure 1 shows the tumor localization by radioimmunoimaging after injection of a mixture of the F(ab')$_2$ fragments as compared with either component admin-

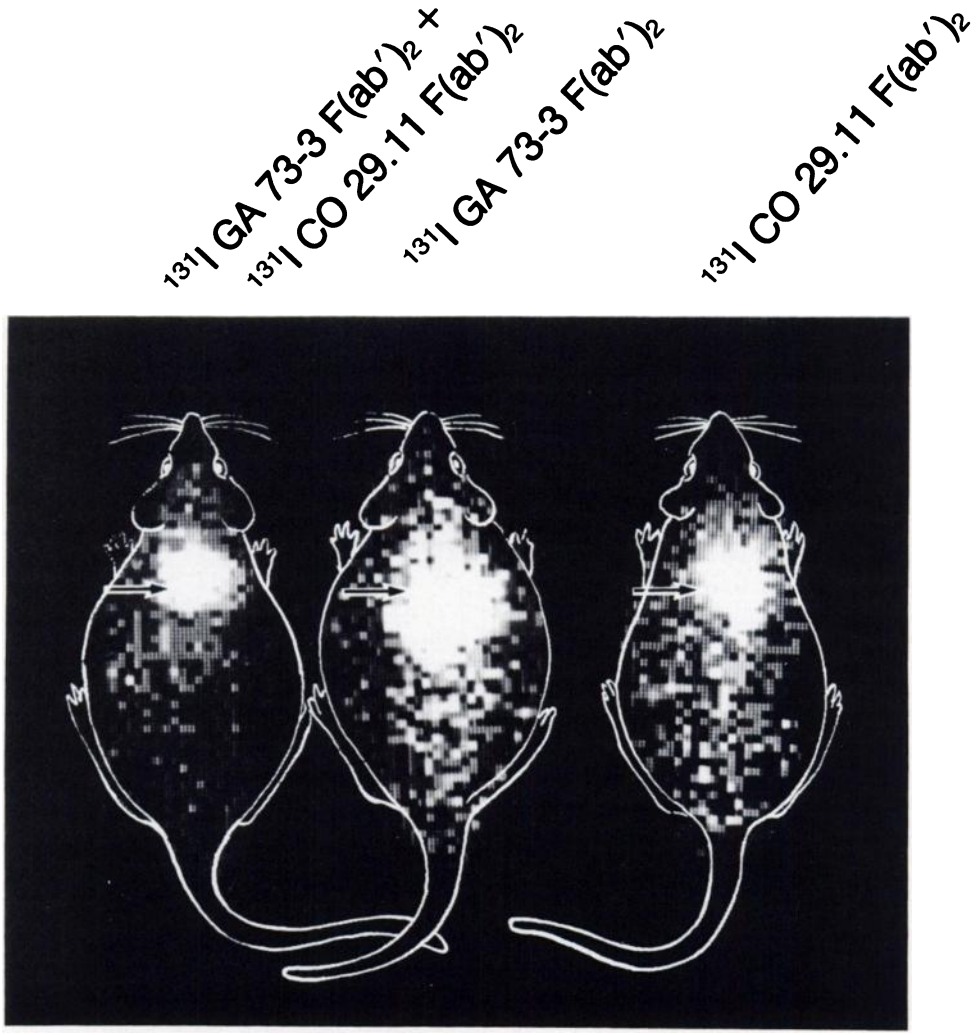

FIGURE 1
Localization of mixture of 100 μCi (11 μg) each of ^{131}I-labeled F(ab')$_2$ fragments of MAbs GA 73-3 and CO 29.11 vs. 100 μCi (11 μg) of either fragment administered alone in nude mice bearing SW-948 CRC tumors (arrows). Posterior view whole-body scans were performed on Day 4 after injection. No background subtraction or computer smoothing was used. Images clearly document the enhancement of tumor contrast using the mixture as compared with either component given separately. Ratios of cpm/pixel in the tumor to rest of the body were 7.1 for F(ab')$_2$ mixture (tumor weight, 100 mg), 3.0 for GA 73-3 F(ab')$_2$ (tumor weight, 130 mg), and 2.8 for CO 29.11 F(ab')$_2$ (tumor weight, 110 mg). Each mouse weighed 22 g

istered separately. Clearly, the tumor contrast obtained with the mixture is a considerable improvement over that using either component alone. This was evident both in posterior and lateral views.

As shown in Table 3, a total of 41 nude mice bearing SW-948 CRC tumors were divided into five groups and injected either with the mixture of ^{131}I-labeled F(ab')$_2$ fragments of MAb GA 73-3 and CO 29.11 or with either fragment alone at various doses. The distribution of tumor weight was similar in all groups of animals examined (30–200 mg). Most of the larger tumors in each group imaged better than the smaller ones. Ratios of cpm/pixel in the tumor and rest of the body, representing tumor contrast, were determined for each mouse daily during the entire observation period of 6 days. The peak ratios obtained in each animal were used in calculating the mean values for each group. These values did not differ significantly in the groups of mice given 100 or 200 μCi of fragments of either MAb GA 73-3 or CO 29.11. On the other hand, the values were significantly higher in the group of mice injected with a mixture of 100 μCi of each MAb fragment as compared with the groups given 100 or 200 μCi of either fragment alone ($p < 0.02$ to < 0.002, Mann-Whitney U-test). A mixture of smaller doses of 50 μCi ($\simeq$6 μg) of each MAb F(ab')$_2$ fragments did not enhance tumor contrast as compared with the use of 100 μCi of either component alone (data not shown).

Fragments of MAb GA 73-3 demonstrated a higher specific activity (cpm/pixel) in the tumor as compared with those of CO 29.11. Especially for the larger doses (200 μCi) injected of each antibody, this effect was highly significant ($p < 0.002$, t-test).

Administration of increasing doses (100–200 μCi/mouse) of MAb fragments alone led to a significant increase in the specific activity (cpm/pixel) in the tumor. With the larger dose of MAb GA 73-3, values in the order of those of the MAb mixture were obtained (Table 3). However, a concomitant increase in the activity in the rest of the body up to levels significantly ($p < 0.01$, t-test) higher than those obtained after the administration of the MAb mixture was also noted. Hence, tumor contrast did not increase with increasing doses of MAb fragments given alone, and highest contrast was obtained following the mixture of both antibodies.

Table 3 also summarizes the biologic half-lives ($T_{½}$biol.) in tumor, rest of the body, and total body, determined in the various groups of mice. The $T_{½}$biol. in the CRC tumors after injection of the mixture of the MAb F(ab')$_2$ fragments was significantly longer than the $T_{½}$biol. values determined in the four groups given either MAb fragment alone ($p < 0.01$ to < 0.002, Mann-Whitney U-test). $T_{½}$biol. in the rest of the body was similar in all groups of mice. There was no correlation between tumor size and the corresponding $T_{½}$biol. of the fragments in either tumor or rest of the body.

DISCUSSION

In this paper, we have demonstrated specific localization of colon carcinomas xenografted to nude mice

TABLE 3
Comparative Radioimaging of SW-948 CRC Tumors with ^{131}I F(ab')$_2$ Fragments of MAbs GA 73-3 and CO 29.11 Administered Separately or in a Mixture

MAb F(ab')$_2$	Radioactivity dose per MAb F(ab')$_2$ (μCi)	Protein dose per MAb F(ab')$_2$ (μg)	Tumor/rest of body ratios of radioactivity [(cpm/pixel in tumor)/(cpm/pixel in rest of body)]	Specific activity in tumor (cpm/pixel)	Specific activity in rest of body (cpm/pixel)	Biologic half-life (hr)		
						Tumor	Rest of body	Total body
GA 73-3† CO 29.11 (n = 9)	100	11	3.88 ± 1.51*	3.01 ± 1.15†	0.85 ± 0.35†	44.7 ± 14.5‡	26.6 ± 3.9‡	38.4 ± 12.5‡
GA 73-3 (n = 8)	100	11	2.33 ± 0.73	1.47 ± 0.85	0.67 ± 0.20	29.6 ± 8.1	25.7 ± 4.7	26.7 ± 5.2
GA 73-3 (n = 8)	200	25	2.26 ± 0.31	3.14 ± 0.77	1.44 ± 0.23	26.4 ± 2.4	24.1 ± 1.8	24.9 ± 2.0
CO 29.11 (n = 8)	100	11	2.02 ± 0.40	1.16 ± 0.45	0.60 ± 0.17	23.0 ± 7.5	19.7 ± 3.7	20.6 ± 2.0
CO 29.11 (n = 8)	200	26	1.92 ± 0.17	1.85 ± 0.85	1.03 ± 0.20	26.8 ± 2.5	24.3 ± 0.7	24.7 ± 0.7

*Mean ± s.d.; peak ratios obtained in each mouse during 6-day observation period were used in calculating mean values for each group.
†Mean ± s.d. determined at Day 4 after injection.
‡Mean ± s.d.

by ^{131}I-labeled (Fab')$_2$ fragments of MAbs GA 73-3 and CO 29.11 and enhancement of tumor contrast on radioimmunoscans using a mixture of these fragments as compared to either component administered alone. Imaging of tumors was successful without using subtraction methods and using MAbs against shed as well as nonshed antigens, thereby confirming previous results obtained in our laboratory (*6,7*). Accumulation of F(ab')$_2$ fragments of GA 73-3 in CRC tumors in vivo was higher as compared with that of CO 29.11, reflecting the higher immunoreactivity of iodinated GA 73-3 F(ab')$_2$, demonstrated here and/or the higher antibody affinity and antigen density on the tumor cell surface. Regarding the latter hypothesis, a statistically significant correlation has been demonstrated between the products of the two binding parameters (association constant Ka times number of antibody binding sites per cell) and the ability of radiolabeled F(ab')$_2$ fragments to localize tumor grafts in vivo (*19*). For MAb GA 73-3, the products is 4.5 as compared with 1.5 for MAb CO 29.11 (*6,7*).

Our study clearly documents the improvement of tumor contrast when a mixture of the two MAb F(ab')$_2$ fragments was used. Furthermore, increasing protein doses of either MAb alone beyond the total dose used in the mixture did not yield tumor contrast comparable to that obtained with the mixture of both MAbs.

The enhanced tumor contrast after administration of the mixture most likely reflects binding of MAbs GA 73-3 and CO 29.11 to distinct antigenic determinants (*6,7*), resulting in increased total MAb binding. Moreover, the increase is likely in radioactivity bound per cell rather than in percentage of cells labeled, since MAbs GA 73-3 and CO 29.11 bind to 100% and 60% of SW-948 CRC cells, respectively (*6,7*). These data also suggest that the antigenic sites detected by MAbs GA 73-3 and CO 29.11 on an individual cell are located such that binding of the two MAbs is not subject to steric interference.

The longer biologic half-life of the mixture of both MAb fragments in the tumor as compared with either component alone might further contribute to the enhancement of tumor contrast, although the mechanisms responsible for this effect are unclear. Nevertheless, this notion is attractive insofar as it suggests the use of mixtures of both ^{131}I-labeled F(ab')$_2$ MAbs in radioimmunotherapy.

In light of the heterogeneity in the expression of antigenic determinants not only within single tumors of a patient but also among different primary and metastatic lesions (*20*), application of mixtures of MAb F(ab')$_2$ fragments directed against different antigens might result in both enhanced tumor contrast and/or detection of more tumor sites. MAb mixtures consisting of more than two components might yield even better results, and are preferable to the use of polyclonal reagents in light of the better tumor selectivity and specificity as well as reproducibility of the MAb preparation.

Iodine-131-labeled F(ab')$_2$ fragments of MAbs GA 73-3 and CO 29.11 seem likely candidates for clinical diagnostic and therapeutic trials, especially if used as mixtures.

FOOTNOTES

* Centocor Inc., Malvern, PA.
† Pierce Chemical Co., Rockford, IL.
‡ Radiochemical Center, Amersham, England.
§ Bio-Rad Laboratories, Richmond, CA.
¶ Siemens Medical Systems, Inc., Des Plaines, IL (Phogamma LFOV).
** Digital Equipment Corp., Maynard, MA (PDP 11).

ACKNOWLEDGMENTS

This work was supported by a grant from the Deutsche Forschungsgemeinschaft, Mu-693/1-1, and National Institutes of Health grants CA-10815, CA-21124, and CA-33491. We thank Mehran Khorsandi for his help in statistical analysis of the data and Marina Hoffman for editorial assistance.

Dieter Ludwig Munz was supported by the Deutsche Forschungsgemeinschaft (Heisenberg-Program), West Germany.

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