Preclinical Assessments of 90Y-labeled C110 Anti-Carcinoembryonic Antigen Immunotoxin: A Therapeutic Immunoconjugate for Human Colon Cancer

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ABSTRACT

We have synthesized 90Y-labeled immunotoxin (IT) containing ricin A chain and C110 anti-carcinoembryonic antigen monoclonal antibody (MAb) to produce a therapeutic immunoconjugate for human colon cancer. The C110 IT was labeled with 90Y via a benzylisothiocyanate derivative of diethylenetriaminepentaacetic acid. The efficiency of 90Y labeling was consistently 90 to 98%, with a specific activity of about 1 μCi/μg. In in vitro stability studies, more than 80% of 90Y remained bound to the C110 IT for up to 5 days after incubation. The percentage of binding of 90Y-labeled C110 IT to carcinoembryonic antigen-coated microbeads was 86%, indicating good retention of the initial immunoreactivity of the C110 MAb. In in vitro protein synthesis inhibition assays, 90Y-labeled C110 IT was approximately 3.7-fold more toxic to the LS174T human colon carcinoma cell line than unmodified C110 IT and 1380-fold more toxic than 90Y-labeled C110 MAb. Biodistribution studies of 90Y-labeled C110 IT in LS174T tumor-bearing mice showed that, at 24 h following i.p. injection, high accumulation of radioactivity was seen in the i.p. tumor and liver and, thereafter, high accumulation in these tissues remained almost unchanged until up to 168 h, with percentage of injected dose/g ranging from 15 to 18% in the tumor and 10 to 15% in the liver. The radioactivity in the spleen and bone gradually increased with time and reached their highest levels (approximately 8% of injected dose/g) at 168 h. Estimation of absorbed radiation doses to the tissues showed that i.p. tumor would have received an approximately 1.5 to 7 times higher radiation dose than normal organs. In vivo therapeutic trials, 90Y-labeled C110 IT provided survival prolongation of LS174T tumor-bearing mice superior to that with either unmodified C110 IT or 90Y-labeled C110 MAb (p < 0.01; Mann-Whitney U test).

These results indicate that 90Y-labeled C110 anti-carcinoembryonic antigen IT may be a potent therapeutic immunoconjugate for human colon cancer and that it may have direct relevance for i.p. treatment of peritoneal carcinomatosis from colon cancers.

INTRODUCTION

Targeted cancer therapy using immunoconjugates, MAbs coupled to the various kinds of cytotoxic agents, is receiving intense scrutiny in current cancer therapy research. Among these immunoconjugates, ITs containing ricin A chain and MAbs have shown selective cytotoxicity in vitro for target cells (1-10) and antitumor activity in vivo in tumor-bearing mice (11, 12). The cytotoxic activity of ITs is based upon the internalization of ricin A chain into the cytoplasm, which results in inhibition of ribosomal protein synthesis. Large molecules such as ITs must be internalized in a cancer cell to cause cell killing.

Thus, incomplete internalization of ITs in all of the cancer cells may limit the effectiveness of this form of therapy. Since MAbs labeled with therapeutic radionuclides produce cytotoxic effects by their emitted high energy particles, internalization of the radionuclides is not necessary for radiolabeled MAbs to produce their cytotoxic effects. In addition to the cytotoxic effects on labeled cells, adjacent cells with or without surface antigen expression also can be damaged. Therefore, radiolabeling of ITs with therapeutic radionuclides may augment the effectiveness of ITs and overcome the problem of ITs described above, thereby providing more potent reagents for cancer therapy than ITs or radiolabeled MAbs alone. For this purpose, we have synthesized an IT containing ricin A chain and C110 anti-CEA MAb, further labeled with 90Y, which is one of the radionuclides being used in radioimmunotherapy (13).

In this paper, we describe our preclinical assessments of 90Y-labeled C110 anti-CEA IT, as compared with unmodified C110 IT and 90Y-labeled C110 MAb. The results obtained in this study indicate that 90Y-labeled C110 IT may be a potent therapeutic immunoconjugate for human colon cancer.

MATERIALS AND METHODS

Monoclonal Antibody and Immunotoxin. The C110 MAb (IgG1) directed against CEA was obtained from Abbott Laboratories (Abbott Park, IL) as a MAb-DTPA conjugate. The production of C110 MAB and its coupling to the benzylisothiocyanate derivative of DTPA (14) have been described by Sumerdon et al. (15). The average number of chelating groups incorporated into each antibody, determined by a 57Co binding assay (16), was 4 DTPA groups/antibody molecule.

C110 IT-DTPA conjugates were prepared in our laboratory by coupling native ricin A chain (Inland Laboratories, Austin, TX) to the C110 MAB-DTPA conjugate via disulfide linkage, as described previously (7, 8). Briefly, C110 MAB-DTPA conjugates were reacted with a 15- to 20-fold molar excess of N-succinimidyl-3-(2-pyridyldithio)propionate reagent (Pharmacia, Piscataway, NJ) (17). The reaction proceeded at room temperature for 30 min, after which time reaction products were dialyzed against PBS for 16 h at 4°C. The N-succinimidyl-3-(2-pyridyldithio)propionate-derivatized C110 MAB-DTPA conjugates were then reacted with a 5-fold molar excess of ricin A chain overnight at room temperature. The resulting C110 IT-DTPA conjugates were purified by gel filtration on a Sephadex G-200 column and then analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (18). The immunoreactivity of C110 IT-DTPA conjugate was assessed by an enzyme linked immunosorbent assay, and immunoreactivity comparable to that of the underivatized C110 MAB was shown with this immunoconjugate (tested by Abbott Laboratories).

Labeling of C110 IT-DTPA and MAB-DTPA Conjugates with 90Y. The 90Y used in this study was obtained from an in-house 90Sr/90Y radionuclide generator developed in our laboratory. The construction of the generator, its elution, and the conversion of 90Y-EDTA to the acetate complex have been described by Hnatowich et al. (19, 20).

Before labeling with 90Y, the C110 IT-DTPA conjugate at 1 mg/ml was dialyzed against 4 liters of 6.0 mM DTPA-50 mM Na2CO3-0.9% NaCl-trace metal-free solution, pH 8.0, at 4°C, using a sulfur-free EDTA-treated cellulose dialysis tube (Spectrum, Los Angeles, CA), for 2 days. The conjugate was then dialyzed against 50 mM Na2CO3-0.9%
NaCl-TMF solution, pH 8.0, at 4°C, to remove free DTPA. C110 IT-DTPA and MAb-DTPA conjugates were mixed with carrier-free $^{90}Y$-acetate and incubated at room temperature for 30 min. Labeling efficiency in the final samples was determined by ITLC (21). Briefly, $^{90}Y$-labeled C110 IT or MAb was incubated with 0.05 M DTPA, pH 6.0, at room temperature for 10 min. A 3-$\mu$l aliquot of each sample was spotted onto an ITLC-SG strip (1 x 8 cm; Gelman Sciences, Ann Arbor, MI). The ITLC strips were developed with 0.9% NaCl solution as solvent. In this system, protein-bound $^{90}Y$ remains at the origin of the strip, whereas free $^{90}Y$ moves with the solvent front. After the solvent moved to the top of the strip, the strip was dried, cut in half, and counted in a gamma counter (Packard Instrument, Downers Grove, IL), with the energy window set to 50 to 500 keV. The percentage of applied $^{90}Y$ bound to the protein was calculated by comparing the net cpm of the origin section of the strip with that of both origin and solvent front sections.

To determine the in vitro stability of incorporated $^{90}Y$, $^{90}Y$-labeled C110 IT or MAb was incubated in human serum at 37°C. The percentage of $^{90}Y$ radioactivity retained in the IT or MAb was measured by ITLC over a period of 5 days.

Immunoreactivity of $^{90}Y$-labeled C110 IT and MAb was determined by a binding assay using CEA-coated Sepharose beads (15). Briefly, a 50-$\mu$l aliquot of $^{90}Y$-labeled C110 IT or MAb, previously diluted with PBS to a conjugate concentration of 0.5 µCi/ml, was added to a microtiter plate with 96 wells coated with 50 nM CEA. Following a 1-h incubation at 37°C, the microtiter plates were frozen for 4 weeks to stabilize the antibody-antigen interactions. The beads were washed by resuspension in 2.5 ml of PBS and recentrifugation, with this being repeated three times. The final bead pellet was resuspended in 0.5 ml of PBS and counted again in a gamma counter. The percentage of radiolabeled C110 IT or MAb bound to the CEA-coated beads was determined by comparing the net cpm of the test tube contents after washing with that before washing.

**In Vitro Cytotoxicity Assay.** Cytotoxicity was measured by inhibition of protein synthesis, as described previously (12). Briefly, LS174T human colon carcinoma cells (22), suspended in leucine-free minimum essential medium, were placed into a 96-well flat-bottomed microtiter plate (Dynatech Laboratories, Arlington, VA), 5 x 10^4 cells in a final volume of 200 µl/well. The cells were then incubated with PBS alone (control) or with specific additions of $^{90}Y$-labeled IT (5.1 x 10^-8 M, 5.1 x 10^-9 M, 5.1 x 10^-10 M, or 5.1 x 10^-11 M) for 18 h at 37°C in 5% CO2. Similar experiments were performed with the unmodified C110 IT and $^{90}Y$-labeled C110 MAb. The medium was then removed and replaced with leucine-free minimum essential medium supplemented with [3H]leucine (New England Nuclear, Boston, MA) at a concentration of 1.5 µCi/well in 200 µl. Following a 1-h incubation at 37°C in 5% CO2, the microtiter plates were frozen for 4 weeks to allow for $^{90}Y$ decay. Following a 10-min treatment with 0.05% trypsin-0.02% EDTA (100 µl/well), the cells were harvested onto glass fiber filters using a MASH II cell harvester (Microbiological Associates, Bethesda, MD) to remove any [3H]leucine that was not incorporated into cellular protein. The glass fiber discs were dried, and [3H]leucine incorporation was determined by liquid scintillation counting. The average cpm of the quadruplicate treated cell samples was calculated and compared with that of the untreated control cell samples. The results were expressed as the percentage of control response.

In enhancing studies with monensin (a carboxylic ionophore) (23, 24), the cells were preincubated with 10^-7 M monensin (Calbiochem, La Jolla, CA) for 1 h and then treated with specific additions of $^{90}Y$-labeled C110 IT or unmodified C110 IT (1.6 x 10^-10 M, 1.6 x 10^-9 M, 1.6 x 10^-8 M, or 1.6 x 10^-7 M) for 18 h at 37°C in 5% CO2. Similar experiments were performed with the unmodified C110 IT and $^{90}Y$-labeled C110 MAb. The medium was then removed and replaced with leucine-free minimum essential medium supplemented with [$^{35}S$]leucine (New England Nuclear, Boston, MA) at a concentration of 1.5 µCi/well in 200 µl. Following a 1-h incubation at 37°C in 5% CO2, the microtiter plates were dried, cut in half, and counted in a gamma counter. The percentage of radiolabeled C110 IT or MAb incorporated into cellular protein was calculated by comparing the net cpm of the test tube contents after washing with that before washing.

**In Vivo Therapeutic Trials.** Two weeks after inoculation of tumor cells, the mice were treated with (a) PBS (control), (b) unmodified C110 IT (80 µg), (c) $^{90}Y$-labeled C110 MAb (80 µg with specific activity of radiolabel of 1 µCi/µg), (d) $^{90}Y$-labeled C110 IT (80 µg with specific activity of radiolabel of 1 µCi/µg), or (e) a mixture of b and c. Each mouse received a 1-ml injection of immunonconjugates or PBS i.p. once a week for 4 weeks. Deaths were monitored daily and recorded when they occurred. Statistical comparison of survival curves was accomplished by the log-rank (Mann-Whitney U) test.

**RESULTS**

Preparation of $^{90}Y$-labeled C110 IT. Analysis of C110 IT-DTPA conjugates by sodium dodecyl sulfate-polyacrylamide gel electrophoresis showed that one to three molecules of ricin A chain were coupled with one molecule of antibody and that the percentage of C110 MAb-DTPA conjugates coupled with ricin A chain was approximately 80%.

The efficiency of labeling of C110 IT-DTPA conjugates with $^{90}Y$ was consistently 90 to 98%, resulting in a specific activity of about 1 µCi/µg. Similar labeling yields were obtained with $^{90}Y$-labeled C110 MAb.

The $^{90}Y$-labeled C110 IT was evaluated for its in vitro stability in human serum by ITLC (Fig. 1). After a 1-day incubation in human serum at 37°C, 93% of $^{90}Y$ was retained in the IT; after a 5-day incubation, 80% of $^{90}Y$ was still associated with the IT. These results were comparable to those for $^{90}Y$-labeled C110 MAb.
The immunoreactivity of 90Y-labeled C110 IT was compared with that of 90Y-labeled C110 MAb by a binding assay using CEA-coated microbeads. The percentage of binding of 90Y-labeled C110 IT and MAb under conditions of approximately infinite antigen excess was 86 and 98%, respectively, indicating excellent retention of the initial immunoreactivity of the C110 MAb during the process of preparing these immunoconjugates. 

**In Vitro Cytotoxicity Assay.** Dose-response curves for inhibition of [3H]leucine incorporation in LS174T cells by 90Y-labeled C110 IT, unmodified C110 IT, and 90Y-labeled C110 MAb are shown in Fig. 2. Both 90Y-labeled C110 IT and unmodified C110 IT had comparable cytotoxic effects on the LS174T cell line and reduced [3H]leucine incorporation in the cellular protein with an IC50 of 2.1 x 10^-6 M and 7.7 x 10^-9 M, respectively. These cytotoxic effects were approximately 130- and 380-fold more potent than that of 90Y-labeled C110 MAb (IC50, 2.9 x 10^-6 M).

Dose-response curves for inhibition of [3H]leucine incorporation in LS174T cells by 90Y-labeled C110 IT and unmodified C110 IT in the presence of monensin (10^-7 M) are shown in Fig. 3. The cytotoxic effects of both ITs on the LS174T cell line were slightly enhanced by 10^-7 M monensin; their IC50 values were 9.0 x 10^-10 M and 1.7 x 10^-9 M, respectively.

**Biodistribution.** The biodistribution of 90Y-labeled C110 IT at various times after i.p. injection into LS174T tumor-bearing mice is summarized in Table 1. The disappearance of radiolabel from the ascites was primarily due to its accumulation in the i.p. tumor and liver and its absorption into the blood at up to 24 h. At 24 h following injection, high accumulation of radiolabel was seen in the tumor and liver, approximately 15 and 10% ID/g, respectively. Thereafter, high accumulation of radiolabel in these tissues remained almost unchanged until up to 168 h. The radioactivity in the blood showed its peak level of 7.28% ID/g at 24 h and thereafter declined with time. The radioactivity levels in the spleen and bone, which were lower than that in the blood at 24 h, gradually increased with time and reached their highest values at 168 h (7.85 and 7.98% ID/g, respectively). The splenic mass in the animals sacrificed at 120 and 168 h decreased by 2- to 3-fold. The radioactivity in other tissues showed peak levels at 24 to 72 h and then decreased when blood radioactivity cleared after 120 h.

The comparative biodistributions of 90Y-labeled C110 IT and MAb in LS174T tumor-bearing mice following i.p. injection were determined by quantitative whole-body autoradiography (Table 2). Representative autoradiograms of the tumor-bearing mice are shown in Figs. 4 and 5. The accumulation of 90Y-labeled C110 IT in the liver was almost 1.5 times more than that of 90Y-labeled C110 MAb at all times. On the other hand, radioactivity of 90Y-labeled C110 IT in the blood decreased more rapidly than that of 90Y-labeled C110 MAb. In the other organs, as well as the tumor, no significant difference was seen between the biodistributions of 90Y-labeled C110 IT and MAb. In addition, the biodistribution of 90Y-labeled C110 IT determined by quantitative autoradiography was almost the same as that determined by tissue counting.

**Radiation Dose Estimates.** Estimates of radiation doses absorbed by the tumor and major organs in LS174T tumor-bearing mice given i.p. injections of 120 µCi of 90Y-labeled...
Table 2 Videodensitometrically determined biodistribution of \(^{90}\text{Y}\)-labeled C110 IT and MAb in LS174T tumor-bearing mice

Mice were inoculated i.p. with \(1.5 \times 10^7\) LS174T cells. Two weeks later, the mice were given i.p. injections of 120 \(\mu\)Ci of \(^{90}\text{Y}\)-labeled C110 IT or MAb (specific activity, 1 \(\mu\)Ci/\(\mu\)g). Radioactivity in tissues was determined by quantitative whole-body autoradiography and is expressed as the percentage of ID/g of tissue at 24, 72, and 120 h following radiolabel injection. The values for percentage of ID/g were normalized to a body weight of 25 g by multiplying by body weight/25. The tabulated values are geometric means of percentage of ID/g from two mice.

<table>
<thead>
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<th>Tissue</th>
<th>(^{90}\text{Y})-labeled C110 IT</th>
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<td>Blood</td>
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Fig. 4. Whole-body autoradiograms of LS174T tumor-bearing mice at 24 h (a), 72 h (b), and 120 h (c) following i.p. injection of \(^{90}\text{Y}\)-labeled C110 IT. Dark areas correspond to high concentrations of radioactivity. Note that the highest concentration of radioactivity is seen in the i.p. tumors at each time point, but a fairly high concentration of radioactivity is also seen in the liver. The radioactivity in the bone increased with time and reached its highest level at 120 h. Arrows, i.p. tumors; Bl, blood; L, liver; B, bone.

C110 IT are shown in Table 3. The doses represent the cumulative radiation doses to the tissues over 168 h and were calculated based on the biodistribution data shown in Table 1. The cumulative radiation dose to the i.p. tumor was calculated as 2780 rad, approximately 1.5 to 7 times greater than those to the normal organs, which range from a high of 1850 rad for the liver to a low of 400 rad for the lungs.

In Vivo Therapeutic Trials. The survival curves of LS174T tumor-bearing mice treated with PBS or various immunoconjugates are shown in Fig. 6. Significant prolongation of survival was achieved in all immunoconjugate-treated groups, in comparison to the control PBS group (\(P < 0.03\) for unmodified C110 IT and \(^{90}\text{Y}\)-labeled C110 MAb; \(P < 0.001\) for \(^{90}\text{Y}\)-labeled C110 IT and a mixture of unmodified C110 IT and \(^{90}\text{Y}\)-labeled C110 MAb). Moreover, \(^{90}\text{Y}\)-labeled C110 IT and a mixture of unmodified C110 IT and \(^{90}\text{Y}\)-labeled C110 MAb provided survival prolongation of LS174T tumor-bearing mice superior to that of either unmodified C110 IT or \(^{90}\text{Y}\)-labeled C110 MAb alone (\(P < 0.01\)). In contrast, survival of mice treated with \(^{90}\text{Y}\)-labeled C110 IT was equivalent to that produced by the mixture of unmodified C110 IT and \(^{90}\text{Y}\)-labeled C110 MAb. In immunoconjugate-treated groups, the cause of all animal deaths was massive progression of i.p. tumors and/or malignant ascites, and no animal deaths were attributable to treatment with the...
designated doses of immunoconjugates or neutropenic infections resulting from myelosuppression by 90Y.

DISCUSSION

The C110 MAb used in this study is an IgG1 murine monoclonal antibody with a high affinity for human CEA. Since specific tumor localization of C110 MAb has been confirmed in the animal (15) and initial clinical studies, this antibody may be considered for its therapeutic application in colon cancer. Our preliminary studies have indicated that conjugates of the C110 MAb with either ricin A chain or 90Y prolonged the survival of nude mice bearing human colon cancer xenografts as i.p. tumors. Thus, we designed this study to assess an immunoconjugate containing both ricin A chain and 90Y on the same C110 MAb molecule as a more potent reagent for colon cancer therapy.

In in vitro PSI assays, 90Y-labeled C110 IT was only 3.7-fold more toxic to the LS174T human colon carcinoma cell line than unmodified C110 IT (Fig. 2). This result may suggest that the radionuclide coupled to IT does not sufficiently enhance IT cytotoxicity even if it is a therapeutic one, such as 90Y or 131I. Buchsbaum et al. (28) have shown a similar result using 90Y-labeled C110 IT in the CEM T-cell acute lymphoblastic leukemia cell line. Since radiation does not affect protein production directly like IT does, the PSI assay may not be a good indication of radiation cytotoxicity from the therapeutic radionuclide. The mechanism of cell killing by radiation probably involves direct or indirect effects on DNA or the plasma membrane. They affect cell proliferation and result in reproductive or interphase death of cells (29). Therefore, alteration in protein production by radiation may not be detectable in the short time period of the PSI assay, which was only 18 h in this study. More potent effects from the radiation may occur in the tumor after longer treatment period.

In vivo therapeutic trials can provide an important method to evaluate potential antitumor effects over a long time course. Our therapeutic trials showed that 90Y-labeled C110 IT produced significantly increased prolongation of the survival of LS174T tumor-bearing mice, in comparison to unmodified C110 IT or 90Y-labeled C110 MAb (Fig. 6). These results suggest that radiolabeling of IT with therapeutic radionuclide may produce additional antitumor activity to that of IT and also that the presence of two cytotoxic agents (i.e., ricin A chain and radionuclide) on the same MAB molecule may provide a more effective and potent reagent for cancer therapy than either agent alone. This additional antitumor activity may be due to (a) killing of the cells that are resistant to IT which do not internalize ricin A chain or do not have surface antigen expression or (b) killing of the adjacent cells that are not bound to IT, by the emitted high energy particles from the radionuclide. The results from in vivo therapeutic trials also suggest that a survival study over a long time course is a good indication of radiation cytotoxicity from the therapeutic radionuclides, which may not be assessed in the short time period of an in vitro PSI assay.

In our therapeutic trials, all animals treated with 90Y-labeled C110 IT died of massive progression of i.p. tumors and/or malignant ascites. No animal deaths were attributable to treatment with the designated dose of 90Y-labeled C110 IT or neutropenic infections resulting from myelosuppression by 90Y. However, in the biodistribution study of 90Y-labeled C110 IT, splenic hypoplasia was observed in animals at 120 and 168 h following i.p. injection. Thus, dose-related toxicity to the hematopoietic organs, which has been reported in treatments using 90Y-labeled MAB (30-33), should also be present in treatments using 90Y-labeled C110 IT, although we did not perform any histological examinations or hematological measurements in this study. Our observations that there were no animal deaths attributable to treatment might be due to the i.p. route of administration. Esteban and colleagues (31, 33) also have reported that i.p. injection of 90Y-labeled anti-CEA MAB (ZCE025) at up to 160 µCi did not cause any radiation-induced mortality, although histological examinations of all treated animals disclosed up to 85% reduction of their bone marrow population and splenic hypoplasia with fibrosis.

The biodistributions of 90Y-labeled C110 IT and MAB in LS174T tumor-bearing mice following i.p. injection were almost identical to each other in the majority of tissues (Table 2). Both immunoconjugates showed specific tumor localization. The only organ in which the biodistributions of 90Y-labeled C110 IT and MAB differed from each other was liver. The accumulation of 90Y-labeled C110 IT in the liver was almost 1.5 times more than that of 90Y-labeled C110 MAB at all times. This higher uptake of 90Y-labeled C110 IT in the liver is attributed to recognition of the mannose and fucose residues on the ricin A chain moiety of the IT by the receptors present on the parenchymal and nonparenchymal liver cells (34). However, chemical destruction of the mannose and fucose residues on the ricin A chain (i.e., deglycosylation) can reduce the liver uptake of ricin A chain (35, 36). In addition, mannose-containing blocking agents, such as ovalbumin, ovomucid, or manno-
syl-lysine, or excess D-mannose or L-fucose also can inhibit uptake of this immunoconjugate by the liver could be eliminated in tumor-bearing mice superior to that of either unmodified C110 IT or 9Y-labeled C110 IT alone. Therefore, the methods described above may reduce the greater uptake of 9Y-labeled C110 IT in the liver.

Whole-body autoradiograms of LS174T tumor-bearing mice in this study revealed that 9Y-labeled C110 IT and MAB injected i.p. were liable to localize to the periphery of the tumor, with little penetration to the center of the masses (Figs. 4 and 5). Microdosimetry within the tumor foci by videodensitometric analysis of the autoradiograms has shown that 9Y-labeled C110 IT and MAB injected i.p. into LS174T tumor-bearing mice may not penetrate into i.p. inoculated tumor foci to a great extent and that the degree of their penetration may not exceed about 1 mm from the tumor surface (data not shown).

In conclusion, immunoconjugates containing both biological toxin and therapeutic radionuclide on the same MAB molecules (i.e., radioimmunotoxins) may be more potent than those containing either agent alone. In this study, we have synthesized an immunoconjugate linked with ricin A chain to the C110 anti-CEA MAB, further labeled with 9Y. This immunoconjugate showed satisfactory efficiency of 9Y labeling, retention of the initial immunoreactivity, in vitro stability, and in vivo cytotoxicity. In vivo therapeutic trials, moreover, showed that 9Y-labeled C110 IT provided survival prolongation of the tumor-bearing mice superior to that of either unmodified C110 IT or 9Y-labeled C110 MAB alone. Therefore, if the greater uptake of this immunoconjugate by the liver could be eliminated using appropriate approaches, it may be a potent therapeutic immunoconjugate for human colon cancer.

REFERENCES


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