Comparison of Indium-111-DTPA-IgG and Tc-99m-MIBI Accumulation in VX-2 Cancer

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ABSTRACT Although Indium-111-DTPA-IgG (In-111-DTPA-IgG) was originally developed as an imaging agent for inflammation/infection, it is known to accumulate in some malignant tumors. Whether such accumulation is related to an inflammatory response to the tumor or to some other mechanism remains to be determined. To address this issue, the accumulation of In-111-DTPA-IgG and Tc-99m-MIBI, which has been recently reported as a tumor-seeking agent, was compared in rabbit VX-2 cancer. In-111-DTPA-IgG and Tc-99m-MIBI were injected into the femoral region of six VX-2 tumor-bearing rabbits 11, 18, and 21 days after implantation of the tumor. On Tc-99m-MIBI scintigraphy, necrosis was visualized as a defect, whereas In-111-DTPA-IgG accumulated in necrotic regions. The In-111-DTPA-IgG or Tc-99m-MIBI tumor to muscle ratios increased as the tumors grew. In-111-DTPA-IgG demonstrated a higher tumor to muscle ratio than Tc-99m-MIBI. In addition, the accumulation of In-111-DTPA-IgG was much higher in necrotic regions than in the viable tumor. These results suggest that the higher tumor uptake of In-111-DTPA-IgG might be mainly accounted for by accumulation in the necrosis and inflammation, whereas Tc-99m-MIBI accumulates in viable tumor cells alone.

Key words: tumor scintigraphy — In-111-DTPA-IgG — Tc-99m-MIBI — VX-2 cancer

Although, to date, a large number of tumor-seeking radiopharmaceuticals have been developed, none of them have proven superior to Ga-67-citrate or Tl-201-Cl in clinical application. In 1988, Rubin *et al*[1] reported increased accumulation of not only radiolabeled specific monoclonal antibodies but also radiolabeled nonspecific polyclonal antibodies in the infectious lesions of rabbits. Since then, studies using indium diethylenetriamine pentaacetate labeled human immunoglobulin (In-111-DTPA-IgG) as a new scintigraphic agent for the detection of infectious and inflammatory lesions have been performed.[2,3] The mechanism of the accumulation of this agent has been considered to be based on protein leakage caused by increased vascular permeability at the inflammatory site.[4,5] Considered from the viewpoint of this accumulation mechanism, it is assumed that the accumulation of In-111-DTPA-IgG is enhanced in malignant tumors where vascular permeability increases. With this in mind, using a rabbit VX-2 cancer, we evaluated the usefulness of In-111-DTPA-IgG as a tumor-seeking agent, comparing it with

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Tc-99m-hexakis-2-methoxy isobutyl-isonitrile (Tc-99m-MIBI), which was originally developed as a myocardial perfusion imaging agent, but has been noted to be a tumor-seeking radiopharmaceutical.[6,7]

MATERIALS AND METHODS

A. Animal model. Albino rabbits weighing 2.5-3.5 kg were used as experimental animals.

B. Transplantation. The VX-2 tumors, an epidermoid carcinoma derived from the Shope virus, were used for transplantation into the thigh muscles. After the tumors were excised from the rabbits and washed, a phosphate buffer containing penicillin 2000 U/ml was added. Then, the suspension was filtered through gauze, and the filtrated fluid was centrifuged. After that, the tumor cells were isolated and prepared in a 20% cell suspension, and 0.1-0.5 ml of this cell suspension was transplanted into the femoral muscles.

C. Imaging of soft tissue tumor. Tumor imaging was performed 11, 18 and 21 days after transplantation. Scintigraphic imaging was carried out after intravenous infusion of pentobarbital (30 mg/kg body weight). Immediately after imaging with Tc-99m-MIBI, In-111-DTPA-IgG was injected, and followed up to 48 hr. Doses of 148 MBq of Tc-99m-MIBI and 40 MBq of In-111-DTPA-IgG were administered through the ear vein. A scintillation camera with a low energy high resolution collimator was used for Tc-99m, and a middle energy collimator was used for In-111. Images with Tc-99m-MIBI were obtained at 5 min. for early images, and 50 min. for late images, and ones with the In-111-DTPA-IgG were obtained at 24 and 48 hr after the administration.

D. Data analysis. The regions of interest (ROIs) in the tumor and contralateral femoral muscle were set, and the tumor to soft tissue accumulation ratio of each agent was calculated. Immediately after In-111-DTPA-IgG imagings, each rabbit was sacrificed under all anesthesia. Next, tumor size was measured, and the radioactivities of In-111 per gram tissue in the viable tumor region, necrotic region and normal muscular tissue were measured. Then, the tumor/muscle and necrotic region/muscle count ratios were calculated.

These experiments were approved by the Animal Research Committee of Kawasaki Medical School (No.95067, 1995), and conducted according to the "Guide for the Care and Use of Laboratory Animals" of Kawasaki Medical School.

RESULTS

The tumor sizes, a minimum of 2.5×2.5×1.5 and a maximum of 6.0×5.5×5.5 cm are shown in Table 1. All intensive accumulations of Tc-99m-MIBI were noted in all tumors except that of one rabbit (No.1) on the 11th day after implantation with minimal tumor size. On the other hand, all tumors showed marked accumulation of In-111-DTPA-IgG. Representative images from 5 to 50 min. after intravenous injection of Tc-99m-MIBI, and at 24 and 48 hr. after administration of In-111-DTPA-IgG are shown in Fig 1. On serial images, Tc-

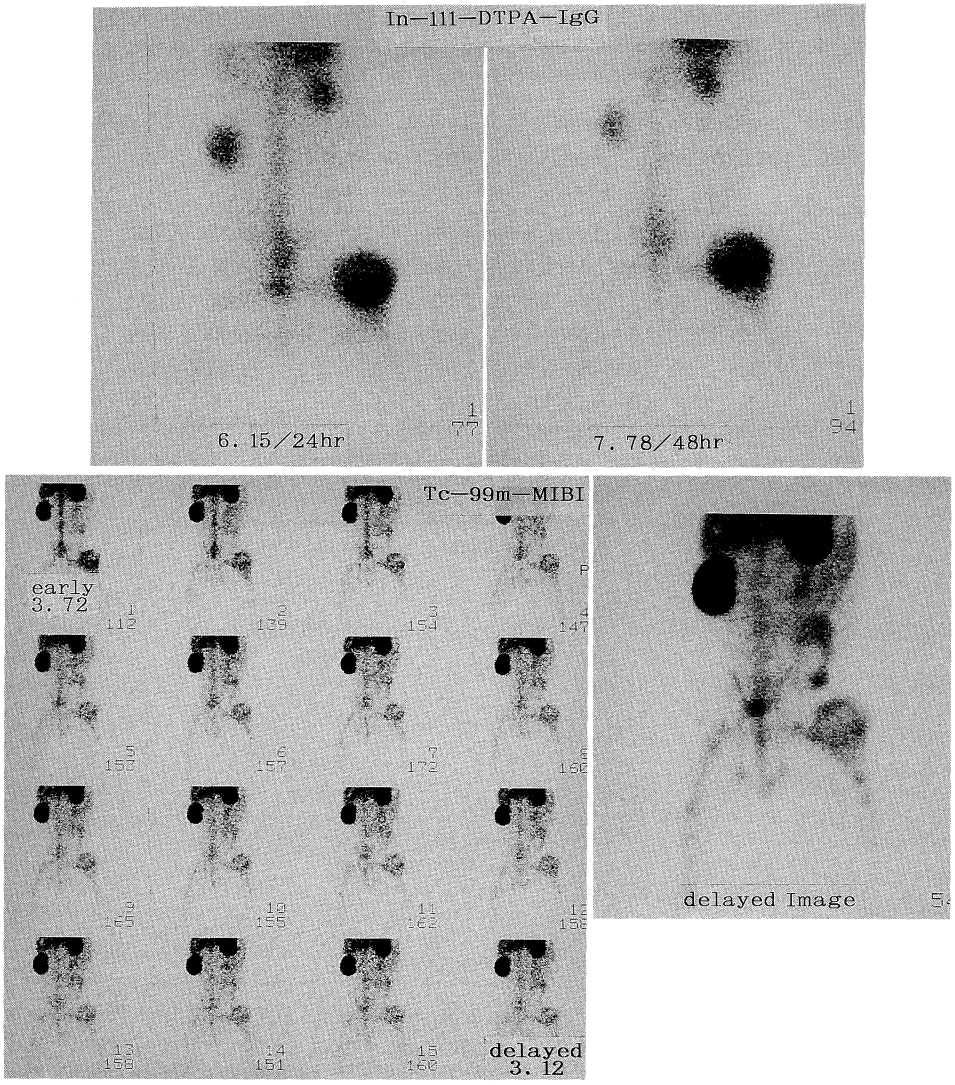

Fig 1. Rabbit No.6 in Table 1.
Tc-99m-MIBI scintigraphy showed increased activity in the tumor region, in which the necrotic region demonstrated decreased uptake. On the other hand, In-111-DTPA-IgG showed intensive accumulation on images at 24 and 48 hr.

99m-MIBI accumulated highly in the tumor region, while decreased uptake was demonstrated in the necrotic region. In-111-DTPA-IgG, on the other hand, showed intensive accumulation on both images at 24 and 48 hr. after injection. In particular, In-111-DTPA-IgG showed significant accumulation in the necrotic region. The tumor uptake ratios of both Tc-99m-MIBI and In-111-DTPA-IgG increased with increase in the size of the VX-2 tumor after implantation. The tumor to soft tissue accumulation ratio was higher for In-111-DTPA-IgG than for Tc-99m-MIBI (Table 1). The counts of In-111 per gram in the tumor region, necrotic region and normal muscular tissue are shown in Table 2. The viable tumor to muscle and the necrotic region to muscle count ratios were higher in the necrotic region than in the tumor.

TABLE 1. Comparison of Tc-99m-MIBI and In-111-DTPA-IgG in tumor to soft tissue accumulation ratio in VX-2 bearing rabbits

Rabbit No.	After implantation (day)	Tumor Size (cm)	Tc-99m-MIBI early (5 min)	Tc-99m-MIBI delayed (50 min)	In-111-DTPA-IgG 24hr	In-111-DTPA-IgG 48hr
1	11 days	2.5×2.5×1.5	1.19	1.02	3.21	3.61
2	11	3.0×2.5×1.5	1.38	1.07	4.76	5.16
3	18	4.5×4.0×2.5	2.12	1.99	4.26	5.24
4	18	5.0×4.0×2.5	3.12	2.35	6.30	7.98
5	21	6.0×5.5×5.5	2.65	2.29	5.02	6.59
6	21	6.0×5.5×5.0	3.72	3.12	6.15	7.78

TABLE 2. Tissue Distribution of In-111-DTPA-IgG in VX-2 bearing rabbits

Rabbit No.	Viable Tumor /Muscle	Necrotic Tissue /Muscle
1	6.30	No necrosis
2	6.13	10.06
3	7.92	12.14
4	10.83	12.26
5	6.24	11.61
6	7.23	16.30

Radioactivity ratio per gram at 48hr postdose

DISCUSSION

Tumor imaging by means of nuclear medicine discloses the functional characteristics of tumors, which are difficult to detect by other morphological imaging methods, such as CT and MRI. For this reason, in addition to the survey of systemic metastatic lesions, much effort has been made in the qualitative assessment of therapeutic effects. To date, a large number of tumor-seeking radiopharmaceuticals have been developed, but none have proven superior to Ga-67-citrate or Tl-201-Cl in clinical application. In recent years, however, Tc-99m-MIBI, developed as a drug for myocardial perfusion imaging, has shown increased accumulation in tumors, and has been applied to tumor imaging.[6,7] In addition to tumor imaging, the development of new radiopharmaceuticals for the detection of infection and inflammatory lesions is also an important goal in field of nuclear medicine. Therefore, much attention is now being focused on In-111 -DTPA-IgG, which might replace Ga-67-citrate or In-111 labeled leukocytes.[8] However, the accumulation of In-111-DTPA-IgG in inflammatory lesions is based on a mechanism of protein leakage caused by enhanced permeability of capillary vessels in the inflammatory process.[4,5] Therefore, it has been proposed that In-111 -DTPA-IgG may also accumulate in malignant tumors where vascular permeability is enhanced.[9] With this in

mind, we carried out a comparative study of In-111-DTPA-IgG and Tc-99m-MIBI as tumor-seeking radiopharmaceuticals in VX-2 tumor-bearing rabbits. As a result, it was found that the tumor accumulation of In-111-DTPA-IgG was more significant than that of Tc-99m-MIBI. With regard to Tc-99m-MIBI, it is considered that its accumulation depends not only on the metabolic activity of the tumor tissue, but also on mitochondria content and such factors as blood flow and enhanced vascular permeability. Tc-99m-MIBI accumulates in cells and is distributed in cytoplasm, primarily mitochondria, depending upon the potential inside and outside the cell or mitochondria membrane. Reflecting the highly active metabolic activity in malignant tumors, the potential differences between mitochondrial membrane might be high, and this may be the cause of increased accumulation of Tc-99m-MIBI in tumors.[10-12] In the case of In-111-DTPA-IgG, on the other hand, it is assumed that accumulation is affected by the leakage of IgG,[4,5] enhanced vascular permeability due to the tumor, or stagnation of IgG or its metabolites in the extravascular space. It also accumulated in the necrotic region regardless of tumor metabolic activity. As for its clinical application, tumor scintigraphy using In-111-DTPA-IgG could prove useful for the detection of tumors, which is difficult to achieve with conventional morphological methods such as CT examination, ultrasonic examination and MRI. However, care should be taken in assessment of the accumulation of In-111-DTPA-IgG in a course after therapy including radiotherapy, as In-111-DTPA-IgG is accumulated not only in the tumor region, but also in the necrotic region. In clinical situations, it is extremely difficult to differentiate the tumor region from nearby inflammation. As In-111-DTPA-IgG shows higher tumor accumulation than Tc-99m-MIBI, and lower physiological accumulation in the abdominal region than Ga-67-citrate, it could prove useful in visualizing tumors of the abdominal or pelvic regions.

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