Radioimmunodetection of Thyroid Carcinoma using Labeled Anti-thyroglobulin Autoantibody

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The word "radioimmunodetection" or "Radioimmunoimaging" means a technique to detect or to present images of tumors using radiolabeled antibodies against tumor specific or tumor-related antigens. Although radioimmunodetection was initially developed to detect coloncarcinoma using radiolabeled antibody against carcinoembryonic antigen (CEA) (1), other antibodies such as anti-alpha fetoprotein (AFP) antibody (2)(3), anti-human chorionic gonadotropin (hCG) antibody (4), antithyroglobulin (Tg) antibody (5) are being employed to detect respectively hepatocellular carcinoma, choriocarcinoma and thyroid carcinoma. In addition, anti-myosin antibody has now been proved to be useful for imaging myocardial infarction.

In the Nagasaki University School of Medicine, we have developed the radioimmunodetection of hepatocellular carcinoma and thyroid carcinoma using respectively anti-AFP and anti-Tg antibodies (2)(3)(5) In this symposium, some recent results on radioimmunodetection of thyroid carcinoma will be shown.

ANIMAL EXPERIMENTS

Materials and Methods

1) Procedures:

Papillary adenocarcinoma, follicular adenoma, Graves' and normal thyroid tissues were transplanted into nude mice. One month later, 25μ Ci of ^{125}I anti-Tg antibody was injected intravenously and scintigrams were taken 3 and 7 days after injection of the antibody. Nude mice were sacrificed after the last scintigram and transplanted tissues and other tissues of nude mice were removed to analyze the ^{125}I-labeled compounds.

2) Preparation of radiolabeled anti-Tg antibody:

Anti-Tg antibody was purified with DEAE Sephacel and affinity chromatography using serum

obtained from a patient with Hashimoto's disease whose anti-Tg antibody titer was 8×10^7 by the Thyroid test. Purified anti-Tg antibody was then iodinated using lactoperoxidase and a glucose-glucose oxidase system.

3) Analysis of radioiodinated compounds in tissues and in sera:

Various tissues were homogenized with a phosphate buffer, and sera and supernatants were filtered through Sephacryl S 300. Each peak of the radioactivity by gel-filtration was then analyzed by affinity chromatography of 1) anti-IgG antibody, 2) anti-Tg antibody, and 3) Tg

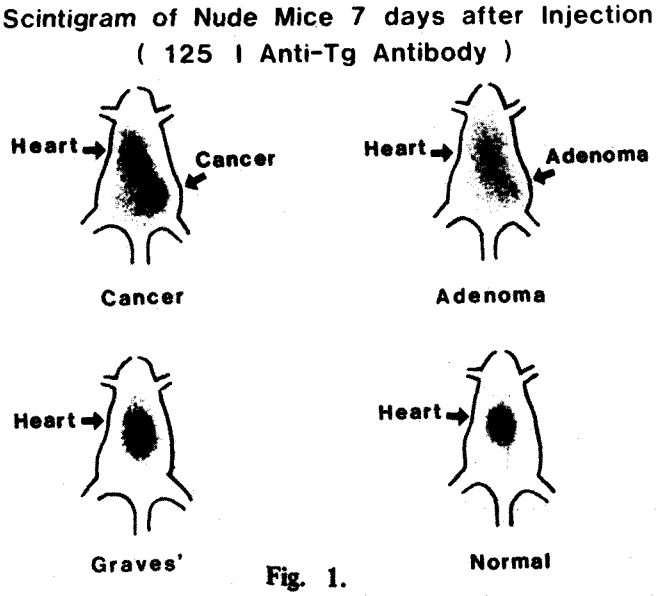

Fig. 1.

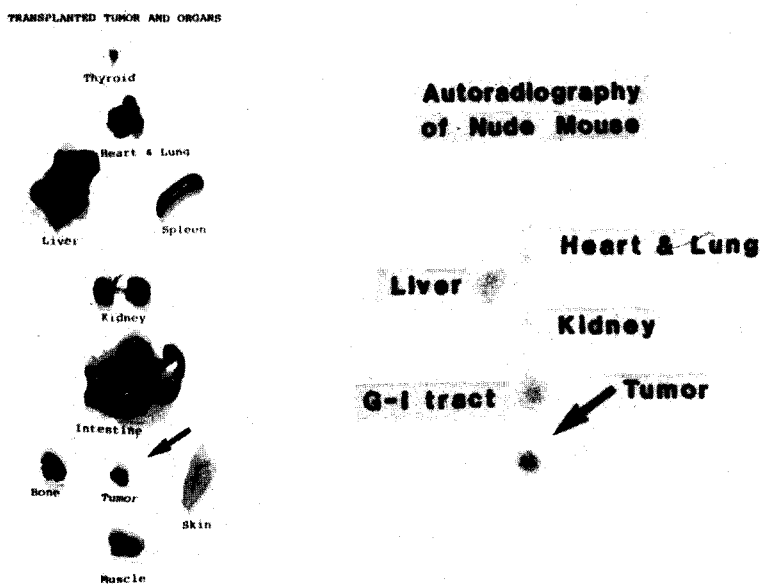

Fig. 2.

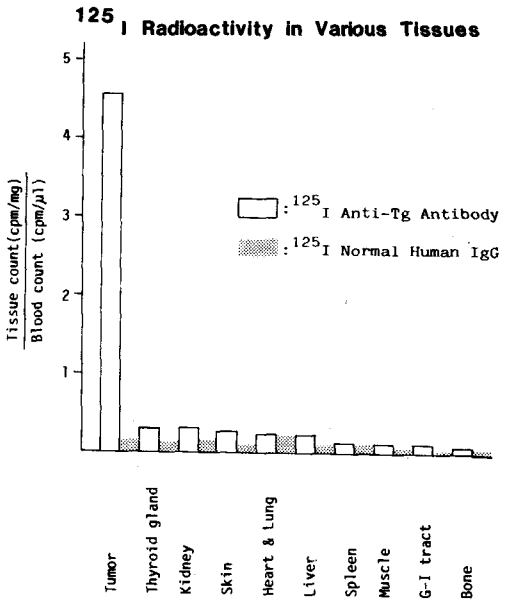

Fig. 3.

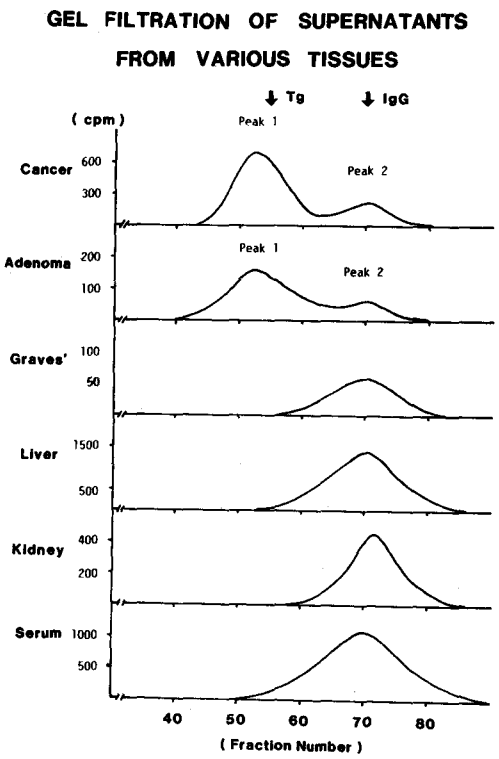

Fig. 4.

bound Sepharose 4B.

Results

Fig. 1 shows the scintigram of nude mice 7 days after injection of ^{125}I anti-Tg antibody. ^{125}I was clearly accumulated by thyroid carcinoma and adenoma tissue transplanted in the right thigh, while Graves' and normal thyroid tissues did not accumulate ^{125}I.

Fig. 2 shows the autoradiography of transplanted tissues and various organs of a nude mouse. As is seen in the figure, transplanted tumor tissue accumulated radioactivity much more than any other organ.

After homogenization of tissues, ^{125}I radioactivity in various tissues was determined in a well type scintillator and the radioactivity in each tissue was expressed as the ratio of the radioactivity of tissue/blood as shown in Fig. 3. ^{125}I was accumulated 4.5 times more in tumor tissues than the blood, while other tissues of nude mice did not accumulate it.

Fig. 4 shows the radioactivity of each fraction of gel-filtration. As seen in the bottom graph, serum samples showed a single peak which is identified as immunoglobulin. Similar peaks were found in many tissues including tumor tissues. The other peak (peak 1) was found only in tumor tissues and aliquots of peak 1 and peak 2 were analyzed with affinity chromatography.

Table 1 shows the results of affinity chromatography analysis. 100% of the radioactivity of peak 2 obtained from serum samples was bound to the anti-IgG antibody and 69.3% was bound to human Tg, indicating that the radioactivity in peak 2 of the serum samples contained radio-

labeled anti-Tg antibody. Since only a very small percent (6.7%) of the radioactivity was bound to the anti-hTg antibody, the radioactivity in the serum did not include an antigen-antibody complex. Similar results were obtained in peak 2 from every tissue tested. However, the radioactivity of peak 1 was also bound to the anti-Tg antibody, indicating that the radioactivity in peak 1 was a Tg and anti-Tg antibody complex.

CLINICAL STUDIES

Since the labeled anti-Tg antibody was accumulated by throid carcinoma or adenoma transplanted in nude mice, an ^{131}I-labeled anti-Tg antibody was injected into patients with carcinoma or adenoma, and scientigrams were taken 3 and 7 days after injection of the antibody.

Fig. 5 shows the scintigram of a patent with an adenoma 7 days after injection of the labeled antibody. In a cold area observed by a routine ^{123}I scintigram, ^{131}I had clearly accumulated. Fig. 6 shows the scintigram of a patient with papillary adenocarcinoma.

There are many methods to detect carcinoma or adenoma within the thyroid. Other methods may be more useful than radioimmunodetection in the diagnosis of thyroid tumors. However, the most useful clinical indication for this technique is a patient with thyroid carcinoma after total thyroidectomy who has high serum thyroglobulin concentrations but metastases are not

Table 1. Affinity Chromatography Analysis

	The supernatant of tumor homogenate		The supernatant of liver homogenate	The supernatant of thyroid homogenate	The supernatant of kidney homogenate	Serum
	peak-1	peak-2				
Anti-IgG-Ab-Sepharose 4B	91.5	100	100	100	85	100
Anti-hTg-Ab-Sepharose 4B	63.5	17.4	6.4	6.8	27.7	6.7
·hTg-Sepharose 4B	32.8	48.8	64.7	81.2	0	69.3

(%)

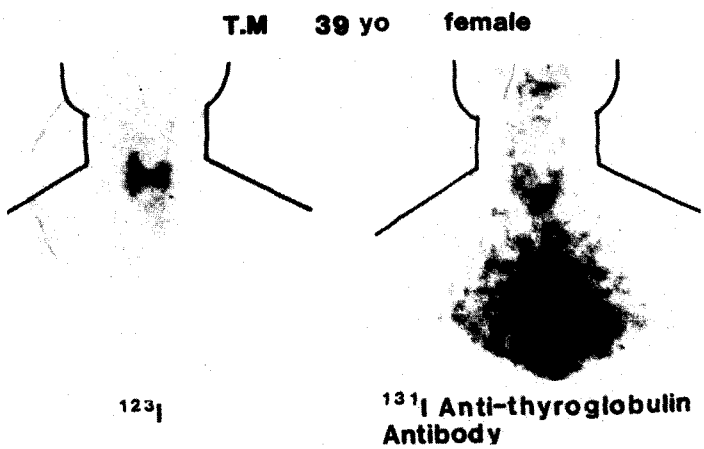

Fig. 5.

detected by the routine ^{123}I or ^{131}I scintigram.

Fig. 7 shows a scintigram of a patient with thyroid papillary carcinoma after total thyroidectomy. Metastasis in the neck lymphnode was not found by any other method. However, after radioimmunodetection, the lymphnode was removed and the diagnosis of metastatic carcinoma was histologically confirmed.

CONCLUSIONS

From these results, it is concluded that the mechanism of Tg secretion in adenoma and carcinoma is different from normal and Graves' thyroids, and radioimmunodetection using an anti-Tg antibody is clinically useful in the diagnosis of thyroid carcinoma.

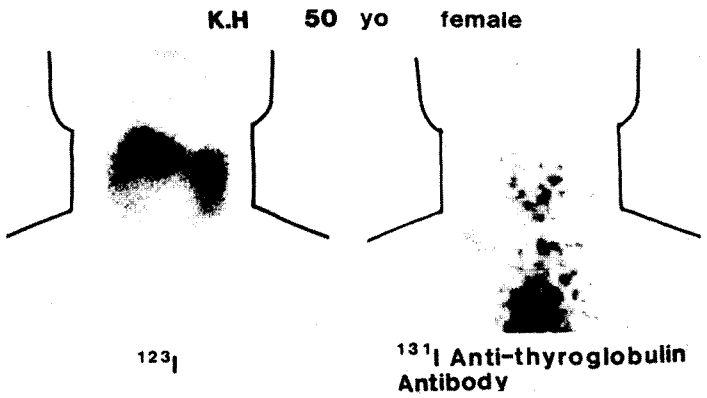

Fig. 6.

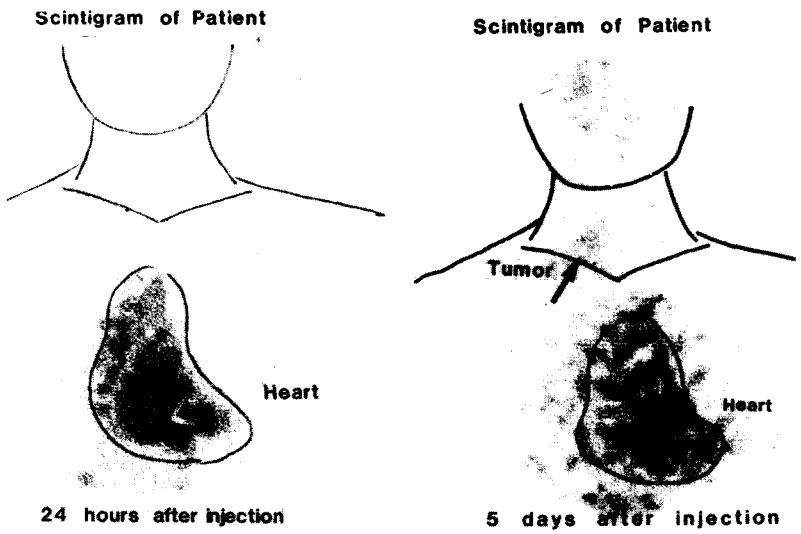

Fig. 7.

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