

102

GAMMACOAT FREE T4 RADIOIMMUNOASSAY. I.Kanamori, S.Matsuo, E.Yasuda, H.Ichikawa, S.Nakano. Ogaki Municipal Hospital. Ogaki.

We have evaluated Japan Travenol's Free T4 RIA kit (GammaCoat 125I Free T4; Clinical Assays, Div. of Travenol Labs, Inc.) for its basic performance and feasibility in diagnosis of thyroid diseases. We have tested the following number of sera from the patients who visited the Ogaki Municipal Hospital; 92 with no detectable symptoms of thyroid diseases, 20 hyperthyroidism, 20 hypothyroidism, 129 normal pregnant, and 18 chronic renal failure.

We have examined/confirmed the following points: 1) Reproducibility of the standard curves (between-assay precision), optimal assay temperature, dilution of specimens, spike and recovery, and the cross-reaction of the antibody-coated tube, all of which have given satisfactory results. 2) Free T4 concentrations by the GammaCoat method are: 1.14 ± 0.24 ng/dl for euthyroid (normal), 3.34 ± 0.27 ng/dl for hyperthyroidism, 0.65 ± 0.42 ng/dl for hypothyroidism. 3) Almost all the pregnant patients have been found to be within the normal range in their free T4 concentrations.

GammaCoat Free T4 Radioimmunoassay requires only a small specimen-size (0.05 ml), short incubation time (20 min + 1 hour), is simple in the principle and the procedure, gives good precision, and satisfies all the basic requirements for routine clinical use.

104

STUDIES ON A SENSITIVE RIA FOR TSH—Applicability Of 200 μ l Serum Sample. S.Bito, Y.Morimoto, H.Ito, N.Oshiro, T.Ishihara, and T.Mori. Nuclear Medicine and Internal Medicine, Kobe Central Municipal Hospital, Kobe.

We have developed a sensitive RIA for TSH which could detect 0.156 μ U/ml by 100 μ l serum sample. (Folia Endocrinol. Jap., 56:1231, 1980) In order to increase the sensitivity further, applicability of 200 μ l serum sample was studied. When Graves' sera were used as carrier, Bo/T by 200 μ l system (15.2—15.6%) were 2 to 2.9% lower than those by 100 μ l system. Other carrier sera—dog serum, horse serum or TSH free human serum (Eiken)—revealed much lower Bo/T and were considered unsuitable as a carrier. By using 200 μ l Graves' serum as the carrier, 0.078 μ U/ml could be detected, and recovery test results of added 0.625 or 2.0 μ U/ml TSH standard to 12 patient sera were excellent as 99.0 and 100.8%, respectively. Assay results of both 100 and 200 μ l system in 83 patient sera showed a good correlation ($r=0.83$), however, 200 μ l assay results of 67 TSH undetectable sera by 100 μ l system were not consistently low but 17 of them even exceeded 0.4 μ U/ml.

Further, standard curves constructed by 4 Graves' sera in 100 μ l system showed quite a good agreement, but those in 200 μ l system were varied widely. Mixing of 2 low TSH sera were resulted in falsely elevated TSH level. In conclusion, in our sensitive RIA system using Daiichi TSH RIA kit application of 200 μ l serum sample was found not effective in improvement of the assay sensitivity.

103

PRELIMINARY STUDY ON THE IMAGING OF THYROID GLNAD USING RADIOLABELED ANTI-THYROID ANTI-BODIES. K.Ikekubo, Y.Iida, K.Kasagi, J.Konishi, K.Torizuka, T.Ishihara and T.Mori. Kyoto Univ. School of Medicine and Kobe Central Municipal Hospital, Kyoto and Kobe.

125 I-labeled anti-rat Tg antibody was injected i.v. into the thyroid-tumor bearing rats that had been received K.I. in water. Rats were killed 48 hrs. after injection and the radioactivity in the various organs was compared. Neither thyroid nor tumor had significant uptake of 125 I-substance. Sucrose density gradient analysis suggests the formation of rTg and anti-rTg immune complex in the circulation. Purification of antibody to thyroid plasma membrane (TPM) from human or rat thyroid gland was performed. Hashimoto's serum IgG or IgG obtained from a rabbit immunized against rat TPM was purified by affinity chromatography on human or rat TPM and labeled with 125 I, respectively. Forty percent of 125 I-labeled antibody bound specifically in vitro to human or rat TPM. However, these labeled preparation also bound to human or rat PM from liver and spleen. After adsorbed with rat liver PM, 125 I-labeled antibody was injected into rats and the distribution of radioactivity was determined in the same procedure as the first anti-rTg experiment. The labeled antibody didn't show significant thyroid localization. Spleen and kidney had an increased uptake of radioactivity.

We concluded that purification of more specific antibody to thyroid PM is required for the imaging of thyroid gland.

105

FUNDAMENTAL AND CLINICAL STADIES ON A IMMO-PHASE TBG RIA KIT. N.Ashida, M.Azukizawa, T.Miki, Y.Kumahara and K.Miyai. Osaka kessei Lab., Dept. of Med. & Geriat. and Dept. of Laboratory Med. Osaka Univ. Med. Sch.

Serum TBG concentrations were measured by a TBG RIA kit (Corning IMMOPHASE). TBG concentrations were calculated by measuring I-125-T4 which are bound to sample TBG-anti TBG complex. Anti-TBG antiserum was supplied as an immobilized form to porous glass beads. T4, in the concentrations of 1-128 μ g/dl, was added to T4 free serum, of which TBG concentrations were 10, 20, and 40 μ g/ml, or to serum of patients with hypothyroidism or pregnancy. Results: Binding of I-125-T4 to TBG-antiTBG complex at 25°C reached plateau at 30 min. The coefficients of variation for intraassay and interassay were 3.8% and 4.8-7.2%, respectively. Fifteen percent less value of TBG concentrations were obtained in the presence of 16 μ g/dl T4 in serum. This influence by serum T4 on TBG measurement was not improved by adding a large amount of BSA or unlabeled T4. Clinical data: Serum TBG concentrations were 17.5 ± 2.7 μ g/ml (mean \pm S.D.) in 15 normal subjects, 43.5 ± 5.6 μ g/ml in pregnant women, and less than 10 μ g/ml in patients with TBG deficiency. TBG values measured by this method tended to be lower in hyperthyroid patients than the values by other TBG RIA kit (Hoechst). T4:TBG ratio employing TBG concentrations by this method correlated well with FT4I ($r=0.989$) or with FT4 concentrations measured by FT4 RIA kit (Corning) ($r=0.975$).