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## RADIOIMMUNOASSAY FOR SERUM FREE THYROXINE USING AMERLEX TOTAL THYROXINE RADIOIMMUNOASSAY SYSTEM.

S. Satake, M. Fukuchi, H. Kitani, M. Murakami, Y. Kaneko, Y. Fujita, M. Hara and K. Nagai. Hyogo College of medicine, Nishinomiya, Hyogo.

Serum free thyroxine (FT<sub>4</sub>) assay system was developed using Amerlex total T<sub>4</sub> kit with minor modification. One ml of 0.9% NaCl was used as a incubation buffer. Standard F T<sub>4</sub> were calculated by equilibrium dialysis method in our laboratory. The following fundamental and clinical data of our system were obtained. The coefficients of variation for two control sera were 5.7-7.0% (inter-assay) and 4.8-6.1% (intra-assay). The normal value for FT<sub>4</sub> ranged 1.04±0.36 ng/dl (mean±S.D.) as determined on 15 healthy adults. Serum F T<sub>4</sub> was increased in all patients with untreated hyperthyroidism (n=10, 3.93±0.76 ng/dl) and decreased in all patients with untreated hypothyroidism (n=10, 0.18±0.09 ng/dl). It was normal in patients with euthyroid thyroid diseases (n=7) and pregnant women (n=8). The coefficients of correlation between our FT<sub>4</sub> values and FT<sub>4</sub> index (T<sub>7</sub>), our FT<sub>4</sub> values and FT<sub>4</sub> values obtained with equilibrium dialysis method, and our FT<sub>4</sub> values and FT<sub>4</sub> values obtained with GammaCoat free T<sub>4</sub> system were r=+0.93, r=+0.78, and r=+0.91, respectively. These data indicate that FT<sub>4</sub> radioimmunoassay system using Amerlex total T<sub>4</sub> radioimmunoassay kit with minor modification was considered quite useful clinically for evaluation of thyroid status.

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EVALUATIONS OF SOLID-PHASE RADIOIMMUNOASSAY WITH AMERLEX PARTICLES FOR SERUM T<sub>3</sub> AND T<sub>4</sub>. K. Ohshima, S. Maruta, I. Kobayashi, H. Ishihara, K. Suwa, S. Kamio and H. Fukuda. Division of Endocrinology, Department of Medicine and First Department of Internal Medicine, School of Medicine, Gunma University, Maebashi.

Usefulness of serum T<sub>3</sub> and T<sub>4</sub> radioimmunoassay (RIA) kit (Amerlex T<sub>3</sub> and T<sub>4</sub>) were evaluated. The antibody-bound fine particles (Amerlex particles) were used as a suspended solution in this solid-phase system. Blood samples were obtained from patients with high, moderate and low concentrations of thyroid hormones. Intra-assay variability of serum T<sub>3</sub> were 1.4 to 4.9%, 2.5 to 5.1% and 4.8 to 6.9%, respectively. Similar data were also obtained with serum T<sub>4</sub>, which were 3.5 to 5.2%, 3.0 to 5.5% and 2.5 to 5.5%, respectively. These results were smaller than those of previously used solid-phase RIA with antibody-coated tubes and similar to those of two antibody method. In addition, inter-assay variability of serum T<sub>3</sub> were 1.7 to 4.0%, while that of serum T<sub>4</sub> were 11 to 16%. The values from this RIA system well reflected the thyroid status with good dilution curves.

In conclusion, measurement of serum T<sub>3</sub> and T<sub>4</sub> by this new RIA system was more rapid than two antibody method, while its reproducibility was similar to the level of two antibody method.

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RADIOIMMUNOASSAY FOR FREE THYROXINE BY MICRODIALYSIS METHOD. K. Kasagi, T. Kosaka, Y. Iida, K. Ikekubo, J. Konishi and K. Torizuka. Kyoto University School of Medicine, Kyoto.

A rapid, simple and accurate radioimmunoassay for FT<sub>4</sub>, Liquisol™ (CIS), has been developed using microencapsulated anti-T<sub>4</sub> antiserum to which <sup>125</sup>I-T<sub>4</sub> has been complexed. The fundamental and clinical evaluation of the RIA kit was performed. Coefficients of variation for 2 control sera were 5.2% and 8.7% (intra-assay) or 7.6% and 11.0% (inter-assay). The percent cross-reactivity of T<sub>3</sub> and rT<sub>3</sub> to the anti-T<sub>4</sub> antibody was 2.6% and 22.4% respectively. Serum FT<sub>4</sub> concentration was 1.61±0.21 ng/100 ml in 32 healthy subjects, 4.70±1.53 in 9 hyperthyroid patients, 0.43±0.09 in 12 hypothyroid patients, 1.57±0.33 in 4 pregnant women, 1.27±0.19 in 3 subjects with TBG deficiency, 1.15±0.30 in 5 patients with liver cirrhosis, 2.03±0.49 in 6 patients with acute hepatitis, 1.03±0.43 in 4 patients with chronic renal failure and 1.47±0.40 in 18 patients with malignant neoplastic disease. Serum FT<sub>4</sub> concentrations measured by CIS FT<sub>4</sub> RIA showed good correlation with FT<sub>4</sub>I (r=0.922), FT<sub>4</sub> by Gamma Coat FT<sub>4</sub> RIA (r=0.913) or FT<sub>4</sub> by equilibrium dialysis (r=0.965). The measurement of FT<sub>4</sub> was clinically useful especially for the diagnosis of subclinical hypothyroidism after radioiodine treatment of Graves' disease and for the follow-up examination on the hypothyroid patients during T<sub>4</sub> replacement therapy.

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EVALUATIONS ON CIRCULATING T<sub>4</sub> DETERMINATION BY SOLID-PHASE RADIOIMMUNOASSAY: COMPARATIVE STUDIES AMONG VARIOUS ASSAY SYSTEMS. S. Hayakawa, K. Isano, Y. Takahashi, N. Yoshida, S. Katakai, K. Ohshima, S. Maruta and I. Kobayashi. R1 Laboratory and Department of Medicine, Maebashi Red Cross Hospital, and Division of Endocrinology, Department of Medicine, Gunma University Hospital, Maebashi.

A solid-phase method was widely employed as BF separation procedures of radioimmunoassay (RIA) system for the hormone. We have measured serum T<sub>4</sub> concentration using several commercially available RIA kits with the solid-phase method. The characteristics of each kit were as follows: A, antibody-coated plastic beads. B and C, antibody-coated polystyrene tubes. D, antibody-coated latex particles. Blood samples were obtained from patients with high, moderate and low concentrations of thyroid hormone and determined 10 times each. Intra-assay variabilities were 6.6, 5.8, 5.3% (A), 7.9, 4.2, 3.9% (B), 3.9, 2.6, 4.0% (C) and 4.3, 4.3, 3.6% (D), respectively. However, some variations of each value among different kits were found.

These observations indicate that normal ranges should be determined by each kit for serum T<sub>4</sub>. On the other hand, excellent correlations of serum T<sub>4</sub> among solid-phase, PEG and two antibody methods were obtained.