D. In Vitro, RIA

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ANALYSIS OF ACCURACY OF DETERMINATION VIEWED FROM PRECISION PROFILE. M.Masaka and T. Yoshimi. Second Dept. of Internal medicine, Hamamatsu Medical College. K.Yamada and M. Kaneko. Dept. of Radiology, Hamamatsu Medical College. S.Sasaki. Olivetti Japan Ltd.

Response error relationship (RER) and precision profile have come to be incorporated in recent years into the control of accuracy of radioimmunoassay. Accuracy of precision profile was analyzed in the present study of 7 items such as IRI, T_4 and T_3 out of all the items routinely determined in our laboratory.

The randomly selected results of 10 times repeated determinations of the standard substances and the results of determinations of the sera under control for blood levels were employed as the test samples. The data were treated on the basis of y=b/(a+x) +c after correction with non-specific binding (NSB).

The slope of RER was within 0.01∿0.03 in all the items evaluated. On the other hand, the results of assessment of accuracy of determinations on the basis of precision profile appreciably vary with the approximation formulae to be used for treatment of data of standard curves. In addition, no consistent inclination can be found out between the accuracy and the correlation coefficient to the theoretical formulae. It was assumed to be useful in evaluating the results of tests of charge of various hormones to grasp the coefficients of variations in various ranges of concentrations.

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RADIOIMMUNOASSAY FOR SERUM h(1-34)PTH BY ADSORPTION WITH SILICAGEL. S.Bannai,M.Aoyama.SRL.N.Yoshiyama.Nakano Universal Hospital.Tokyo.

In order to measure serum bioactive PTH, we tried to radioimmunoassay(RIA)for human (1-34)PTH by adsorption with silicagel. The antiserum to PTH were prepared by immunizing rabbits with a h(1-34)PTH-BSA conjugate. A preparation of labeled PTH was radioiodinated by the chloramine T method. In our method,the final dilution of antiserum was 1:30,000 and 40% binding against h(1-34)PTH was obtained. Immunoreactivity of antibody against bovine PTH was less than 1% cross reactivity. Serum PTH was adsorbed by silicagel,and then eluted it from silicagel was performed by hydrochloric acid-aceton(20:80),and elution was evaporated to dryness at 37°C under a air spray. The completely dried material was solubilized with assay buffer and used in the assay, In the assay,B/F separation was used by final 12.5% PEG. The minimum detectable sensitivity of the assay was 30pg/ml, and 30 normal subjects were less than 120pg/ml. We measured h(1-34)PTH, bovinePTH,Calcium level and Phosphorus level in 38 patients with chronic renal failure before and after hemodialysis. h(1-34)PTH values was 109,2± 72.8pg/ml(M±SD) before hemodialysis; they fell to 53.9±25.7pg/ml(M±SD) after hemodialysis. Measurement of h(1-34)PTH was useful with diagnosis of patients with chronic renal failure and secondary hyperparathyroidism.

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RADIOIMMUNOASSAY OF RAT MALIC ENZYME. T.Mitsuhashi,K.Kubota,N.Kuzuya,H.Ikeda and H.Uchimura. The Third Department of Internal Medicine Faculty of Medicine,University of Tokyo. Tokyo.

Malic enzyme (ME) has been used as a marker enzyme in studying the mechanism of action of thyroid hormones in rat tissue. We intended to establish RIA for ME to achieve higher sensitivity and to measure the enzyme protein itself because photometric assay available was relatively low in its sensitivity.

Purified ME was obtained from rat liver by using affinity chromatography and HPLC. Anti-rat ME antiserum was obtained by immunizing purified ME with Freund's complete adjuvant to rabbits and 1:10,000 diluted antiserum was used for the assay. Iodination was performed by chloramine-T. Incubation time was for 48 h at 4°C and BF separation was done by double antibody method. The assay range was from 1 to 100 ng/tube. A very good correlation was observed between values measured by photometric assay and RIA in rat liver and fat tissues. Using this RIA, ME was detectable in peripheral mononuclear cells in rats.

This RIA is useful to measure ME in small pieces of tissue and to study effects of thyroid hormones on ME in rats.

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CLINICAL STUDIES ON THE MEASUREMENT OF SERUM VITAMIN B_{12} AND FOLATE LEVELS BY VITAMIN B_{12}/FOLATE DUAL RADIOASSAY KIT. Y. Takahara, M. Nakamura, A. Ishibashi, J. Sato, S. Kosuda, Y. Yonahara and M. Kondo. The 2nd Tokyo National Hospital and Medical School of Keio University, Tokyo.

We studied on quantitative determination of vitamin B_{12} and folate in blood using Vitamin B_{12}/Folate Dual Assay Kit (Amersham-Kaken) simultaneously. When the measurement was carried, we should confirm the suitability of its own counter for the discrimination between ^{125}I and ^{57}Co. If the instrument is suitable to count the dual assay the spillover for both isotopes must be less than 3%. Reproducibility, dilution test and recovery rate was good. The serum folate level was found to be higher in hemolysis than in controls, but the serum vitamin B_{12} level was not statistically significant variation. Correlation with Folate Radioassay Kit was r = 0.789, y = 0.87x − 0.26, and that with Vitamin B_{12} Radioassay Kit was r = 0.932, y = 0.84x + 195.65. Serum folate levels ranged from 0.98 to 7.07 (mean 2.63 ± 1.15) ng/ml in normal males and ranged from 0.97 to 6.35 (3.31 ± 1.17) ng/ml in normal females. Serum vitamin B_{12} levels ranged from 291.5 to 995.3 (552.2 ± 144.9) pg/ml in normal males and ranged from 202.9 to 1114.5 (522.5 ± 163.2) pg/ml in normal females.

In certain instances the red cell folate levels may provide a more reliable indicator of folate status.