

after the administration, the anticipation of the final results of ^{131}I treatment for hyperthyroidism is impossible. In cases of recur-

rence, T_3 values were found not so high and re-administration of ^{131}I is not always necessary.

Study on Determination of Free Thyroxine in Serum

T. NAKAGAWA, S. HAMADA, T. MORI, R. MORITA, T. SAKURAI
K. FUJII and K. TORIZUKA

The Second Division, Department of Internal Medicine, Kyoto University, School of Medicine, Kyoto

At present, we do think that the amount of ^{131}I to be given should be kept small and the continuous observation of the patients after the administration is necessary.

It has been generally accepted that free thyroxine in serum is responsible for a thyroid status in the subject. Recently Clark and Horn presented "free thyroxine" index, a factor proportional to the concentration of free thyroxine, and showed that values for this index were closely related to the thyroid status in various thyroid diseases. Our studies confirmed the results indicated previously, and showed that this index was still better than triosorb resin sponge uptake itself.

Further, it was studied to assess the level of free thyroxine by equilibrium dialysis using thyroxine ^{131}I . Tracer dosis of T_4 ^{131}I were added to serum, and incubated at 37°C for 1 hour. There ml. of undiluted serum, containing T_4 ^{131}I were dialyzed against 5–20 ml. of potassium phosphate buffer

(p.H. 7.4, $I = 0.15$) at 37°C for 20–24 hours. Stable thyroxine was added to 3 ml. of dialysate, and was precipitated with 10% MgCl_2 . Radioactivity of the precipitate was determined by a well-type scintillation counter. When 0.02% Merthiolate was added to the outside buffer, the level of free thyroxine was shown to augment with increasing amount of the outside buffer. The same results were obtained in the experiments using further purified thyroxine ^{131}I . However, when Merthiolate was absent in the buffer, the level of free thyroxine did not rise, but rather decreased with increasing amount of the outside buffer. Therefore, it was concluded that Merthiolate increased the level of free thyroxine. Moreover, it was shown that Krebs-Ringer solution diminished the effect of outside volume on the level of free thyroxine, and therefore, it was more suitable than potassium phosphate buffer. The values of normal serum ranged between 0.035 and 0.042.

A New Simple Method for the Determination of Thyroxine in Serum

H. NAKAJIMA, M. KURAMOCHI, T. HORIGUCHI and S. KUBO

Department of Pediatrics, School of Medicine, Chiba University, Chiba

A new simple method to measure serum thyroxine (T_4) was introduced here by using the conventional resin sponge uptake of ^{131}I -triiodothyronine (T_3) (Triosorb test). Different from the conventional resin sponge uptake test, the present method can measure T_4 concentration in the serum, not disturbed

by the amount of T_4 binding protein in the sample serum.

Four ml of 95% ethanol is added to 2 ml of the sample serum and is mixed well. After centrifuging, 4 ml of the ethanol supernatant is dried up in a test tube under nitrogen gas. Then, 1 ml of ^{131}I - T_3 in Tris buffer, and 0.5

ml of the standard pooled serum and 0.5 ml of distilled water are added to this test tube. Eight tenth ml of solution of dried human plasma is available in place of the standard pooled serum. After the material is redissolved completely, this is incubated for 30 min. at 4°C. According to the conventional method, resin sponge uptake is measured by a well-type scintillation counter. T_4 content in the supernatant is determined from the standard curve of $^{131}\text{I}-T_3$ resin uptake values obtained using various concentrations of known T_4 solution; 0.0, 0.25, 0.05, 0.075, 0.10, 0.125, 0.15, 0.20, 0.25, 0.30 and 0.40 $\mu\text{g}/\text{ml}$ of T_4 . A sharp linear increase was seen in the range of T_4 concentration from 0.0 to 0.20 μg . However, its increase became less steep beyond the T_4 content of 0.20 μg . Therefore, the standard curve can be applicable for determining the T_4 concentration of below 20 $\mu\text{g}/100$ ml in serum. Then these calibrated T_4 values are further corrected using the value of recovery of T_4 $^{131}\text{I}-T_4$ from the each sample serum to the ethanol extract. The re-

covery by measuring the same sample obtained a satisfactory reproducibility.

The findings of T_4 iodine ($\mu\text{g}/100$ ml) by the present method in various groups of diseases were: 1) hypothyroidism, 2.3 ± 0.70 (mean \pm SD); 2) euthyroid subjects, 5.7 ± 1.08 ; 3) hyperthyroidism, 10.9 ± 2.30 ; 4) pregnant, 7.0 ± 1.31 ; 5) nephrosis, 2.3 ± 0.94 ; 6) newborn infant, 13.7 ± 2.08 ; and 7) arterial cord blood, 7.1 ± 0.77 . The average PBI values determined simultaneously in the cases with euthyroid, hyperthyroid, hypothyroid, pregnant and nephrotic individuals were 5.9 ± 1.10 , 11.8 ± 1.99 , 2.4 ± 0.58 , 7.7 ± 1.13 , and 2.6 ± 0.70 , respectively. These PBI results were correlated well with the T_4 iodine values.

A satisfactory reproducibility was observed through 35 duplicate determinations. The range of differences of the duplicate analysis was 0–1.9 μg of T_4 iodine/100 ml.

These results proved that this method could be used as the routine clinical diagnostic means in place of PBI.

PVF Sponge and Its Clinical Applications

K. INAGAKI

Tokyo Metropolitan Police Hospital

1. PVF-sponge

PVF sponge here is obtained by acetalization of PVA (polyvinyl alcohol) and is considered to have 1–3 glycol bond in its structure. Maximal degree of the acetalization is theoretically 86.74%.

Swelling with water depends upon the degree of the acetalization and no swelling is observed above 81% of acetalization.

Pores make continuous phase throughout the sponge and are controlled in the manufacturing process.

For obtaining homogeneous product, the most favorable degree of acetalization is considered to be 72%, while 65% sponge was used in this experiment.

2. PVF sponge in clinical use

Acetalized (72%) or semiacetalized (65%) form of PVA was used as absorbant of T_3^x (x . means R.I.-labelled) for the purpose of

thyroid function test.

According to the procedure of Triosorb resin test was the T_3^x -uptake of PVF sponge measured in the following experimental conditions:

1. PVF-sponge (semiacetalized cylindrical piece): 1.5 cm long
2. Temperature: 25°C
3. Incubation time: 60 min.
4. Serum to be tested (twice diluted): 1.0 ml
5. Washing: 7 times (1 sample for 1 min.)

The result of the test proves that this sponge could be available for clinical use, showing normal value between 16 and 24%.

The electrophoretical pattern of T_4^x -added serum on cellulose acetate film stripe was cut along referred to the blackened zone in the inter- α area and the incorporation rate of T_4^x in TBG against the whole T_4^x in value