Radioimmunoassay for Serum T_3

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A specific antibody to T_3 (1-triiodothyronine) was made in rabbit immunization with T_3 conjugated to bovine serum albumin with carbodiimide.

High specific activity (454 mc/mg) of ^{125}I-T_3 was kindly supplied by Dainabot Lab.

A cross reaction of the antibody for T_3 to L-monoiodotyrosine, L-diiodotyrosine, or L-thyroxine was <0.001, <0.001 and 0.16% respectively.

For the determination of serum T_3 values, two methods were examined.

(1) In direct method

Ethanol extraction of thyroid hormone (T_4 and T_3) from test serum (0.1 ml) was dried, then human IgG (2 mg), 32 pg of ^{125}I-T_3 and anti-T_3 antibody (diluted 500 times as a final concentration) were added.

Using the double antibody technique (second antibody; sheep anti-rabbit gamma-globulin), a radioimmunoassay of T_3 capable of detecting the range from 32 to 100 pg has been developed. The slope of the standard curve was abruptly changed between 32 and 600 pg, but became mild from 600 to 1000 pg.

Mean serum concentration in patients were as follows:

euthyroid subjects	0.96 ± 0.52 ng/ml (n = 56)
hyperthyroid	4.15 ± 2.73 ng/ml (n = 17)
hypothyroid	0.49 ± 0.24 ng/ml (n = 21)
Hashimoto's thyroiditis	0.60 ± 0.23 ng/ml (n = 8)
Thyroid cancer	0.81 ± 0.24 ng/ml (n = 4)

(2) Direct method

By addition of salicylic acid (10^{-2} M, a final concentration) in test serum (0.1 ml) to inhibit binding of T_3 to thyroxine binding globulin, serum T_3 value was determined directly. The standard curve of a radioimmunoassay that could be detected the same amounts (range, 32–1000 pg) of T_3, was made using hypothyroid serum that had been removed endogenous T_3 completely by dextran coated charcoal.

Mean serum concentration in patients were as follows:

euthyroid subjects	1.32 ± 0.43 ng/ml (n = 51)
hyperthyroid	7.57 ± 4.49 ng/ml (n = 15)
hypothyroid	0.94 ± 0.44 ng/ml (n = 14)
Hashimoto's thyroiditis	1.15 ± 0.16 ng/ml (n = 5)
Thyroid cancer	1.01 ± 0.14 ng/ml (n = 3)

The determined values by these two methods showed a little difference in various diseases.

Experimental and Clinical Investigation on Thyopac-4 Test, and Thyopac Free Thyroxine

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The results from the experimental investigation were as follows: 1) the standard curve practically delineated as a straight line was found somewhat superiorly convex; 2) Thyopac-4 Value was little affected by various conditions of measurements; 3) Mean difference between the duplicate values was sufficiently small; 4) Examination time of the test a kit was able to