THE EFFECT OF A THYROTROPHIC HORMONE PREPARATION ON THE METABOLISM OF RADIOIODINE IN EUTHYROID, HYPERTHYROID AND ACROMEGALIC INDIVIDUALS [1]

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The role played by the thyroid stimulating hormone (TSH) of the pituitary in thyroid disease has excited speculation for some time. The similarity of the effects produced by this material to some of the physiological changes noted in Graves' disease seems more than coincidental. It was early shown that the injection of a crude anterior pituitary preparation in man would cause an elevation in the basal metabolic rate (1, 2). Later studies have shown that thyrotrophin administration to euthyroid normals will increase the radioiodine uptake of the thyroid and the level of the protein-bound iodine of the serum (3–5). Goldsmith, Stanbury, and Brownell have recently demonstrated that the injection of preparations of this material causes an increase in the rate of release of hormone from the thyroid gland (6). Studies on the rat and chick have demonstrated similar changes (7–10).

Evidence indicates that the circulating hormone of the thyroid is loosely bound to plasma proteins (11). Since this hormone is quantitatively precipitated with the proteins of the plasma (12) it is practicable, following the administration of a dose of radioiodine (I^{131}), to measure the output of thyroid hormone as protein-bound radioiodine. These techniques have been utilized in the present study to determine the effect of a single dose of thyroid-stimulating hormone on the discharge of labeled hormone by the thyroid. The consequences of the injection of this pituitary preparation are compared in euthyroid, hyperthyroid and acromegalic individuals.

METHODS

A test dose of I^{131} (as sodium iodide) of from 100 to 950 microcuries containing not more than 10 micrograms of carrier I^{127} was administered orally. The radioactivity in the serum, in the protein-precipitable fraction of the serum, in the urine, and over the neck was measured daily, and at more frequent intervals following thyrotrophin administration. Chemical measurements of the protein-bound iodine (PBI^{127}) were made using Barker's modification of Chaney's procedure (13). The protein-bound I^{131} was measured by precipitating aliquots of serum with zinc hydroxide in the same tubes in which the total serum activity was measured. Somogyi's method (14) was modified by doubling the strength of all solutions in order to reduce the volumes. Following three washings with distilled water, the radioactivity in the precipitate was determined in a "Texaco" gamma ray cup counter (15) (sensitivity—60,000 counts/minute/microcurie for I^{131}) to a minimum counting accuracy of 2 per cent standard error. Repeat determinations of radioactivity on 10 aliquots of the same serum had a standard deviation of 1.2 per cent of the mean. A standard deviation of 3.1 per cent of the mean was found, however, after these aliquots were precipitated and the radioactivity of the protein-bound I^{131} was determined. Recovery studies demonstrated that exchange between I^{131} and serum was negligible, as has been shown by McConahey, Keating, and Power (16).

The amount of radioactivity over the neck was determined with a well collimated "Texaco" Geiger tube 22 cm. above the suprasternal notch (15). The accuracy of this measurement will depend in part upon the geometry of the thyroid and was found to have a standard deviation of approximately 5 to 10 per cent of the mean in the same patient. The urine specimens were collected in daily 24-hour periods and the radioactivity of an aliquot was assayed by counting with a Geiger tube in a Marinelli beaker (15). All radioactivity measurements were corrected for the resolving time of the equipment involved and for physical decay of the isotope.

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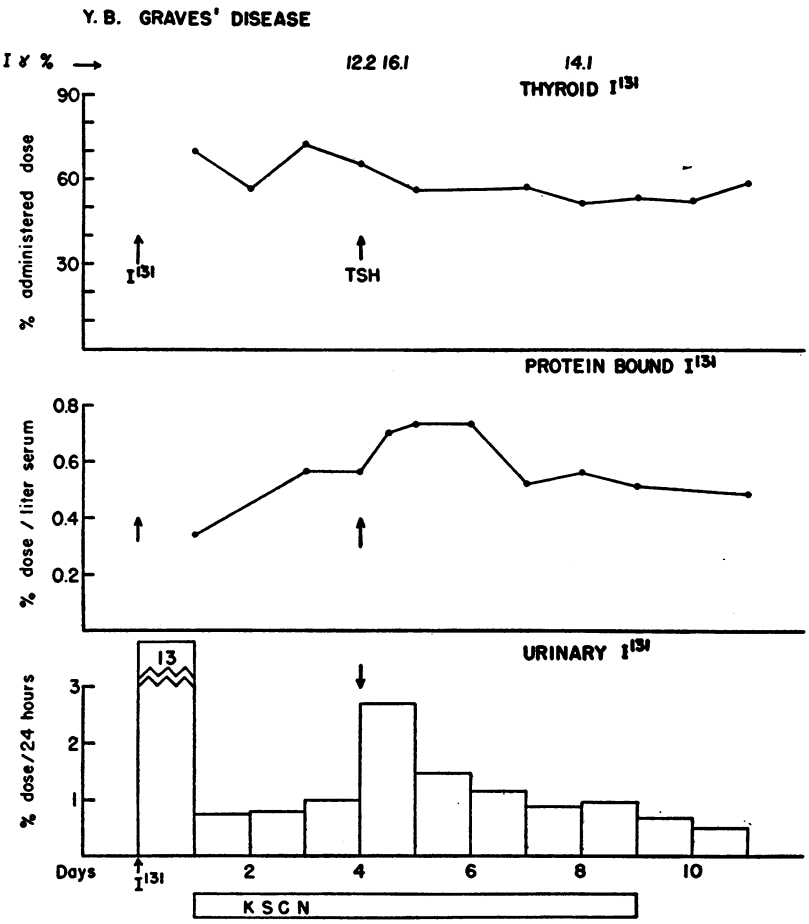

FIG. 1. THE EFFECT OF A SINGLE INJECTION OF TSH ON THE METABOLISM OF I^{131} IN A PATIENT WITH GRAVES' DISEASE

A pituitary preparation[4] (Armour) containing thyroid stimulating hormone was used in this study. The same amount (40 mg. equivalents) of the same preparation was used in 12 of the patients studied. The remaining two (E. G. and B. V.) received 10 mg. equivalents each of the same material. The lyophylized pituitary extract was dissolved before injection in a volume of 2 cubic centimeters of sterile saline and administered intramuscularly.

The experimental subjects were hospitalized (with the exception of S. B., M. L., and S. M.) and kept on a diet relatively low in iodine but otherwise unrestricted.[5] The euthyroid patients were patients with carcinoma of the breast (M. D. and B. M.), arteriosclerotic heart disease (R. B.) and malignant melanoma (F. F.). History, physical examination and the usual laboratory studies (PBI, radioiodine studies, BMR, cholesterol) confirmed their thyroid status. Laboratory examinations demonstrated that all patients had apparently normal renal function. All patients had been under observation in the clinics of either the Memorial or The New York Hospital for some time and their diagnoses were considered to be well established.

The procedure of the experiments was as follows: On the first day, I^{131} was given orally to the fasting pa-

[4] Armour Thyrotropic Factor, Control number J-13903-R. Each vial of this material was stated to contain 20 milligram equivalents of the Armour standard pituitary preparation. Actual weight of the contents of each vial was about 35 milligrams. This material was assayed in the laboratories of the Sloan-Kettering Institute by Dr. William Money to whom we are especially indebted. In this assay, day-old male chicks were injected daily for 3 days with various amounts of this preparation. The animals were killed on the fourth day and the weight of the thyroids determined. Comparison to a standard pituitary preparation assayed in the same manner demonstrated that 1 milligram (actual weight) of the preparation used contained 0.5 Junkman-Schoeller units.

[5] Only uniodized salt was used and foods known to contain large amounts of iodine (mainly sea food) were omitted.

tient. Twenty-four hours later 1 gram of potassium thiocyanate, in the form of enteric-coated tablets, was given orally to prevent the incorporation of further iodine into the thyroid (17). This drug was then continued in a dosage of 0.25 gram twice a day for the duration of the experiment. Serum thiocyanate levels were determined by the method of Gregersen and Stewart (18). When the protein precipitable radioactivity reached a plateau (4 to 8 days after the administration of the tracer) TSH was administered. Determinations were made of serum and neck radioactivity immediately before the injection of TSH and every 2 hours afterward for 8 hours. Eosinophile counts were done in several patients both before and 4 hours after the injection of the TSH.

RESULTS

Figure 1 shows the effects of a single injection of TSH in a patient with Graves' disease. Following the injection of thyrotrophin, no significant change occurred in the readings of radioactivity over the neck. By 24 hours after the injection, however, the PBI^{131} had risen 30 per cent and the PBI^{127} a similar amount.[6] There was a more marked effect on radioactivity measurements in the urine, with the 24-hour collection of urine following the TSH containing over 150 per cent more radioactivity than the 24-hour urine in the period preceding the injection. These same indices are compared in an euthyroid patient in Figure 2.

[6] The changes (in terms of percentage) in the various indices are calculated on the basis of a comparison with the "control" determination—that value immediately preceding the administration of the thyroid-stimulating hormone.

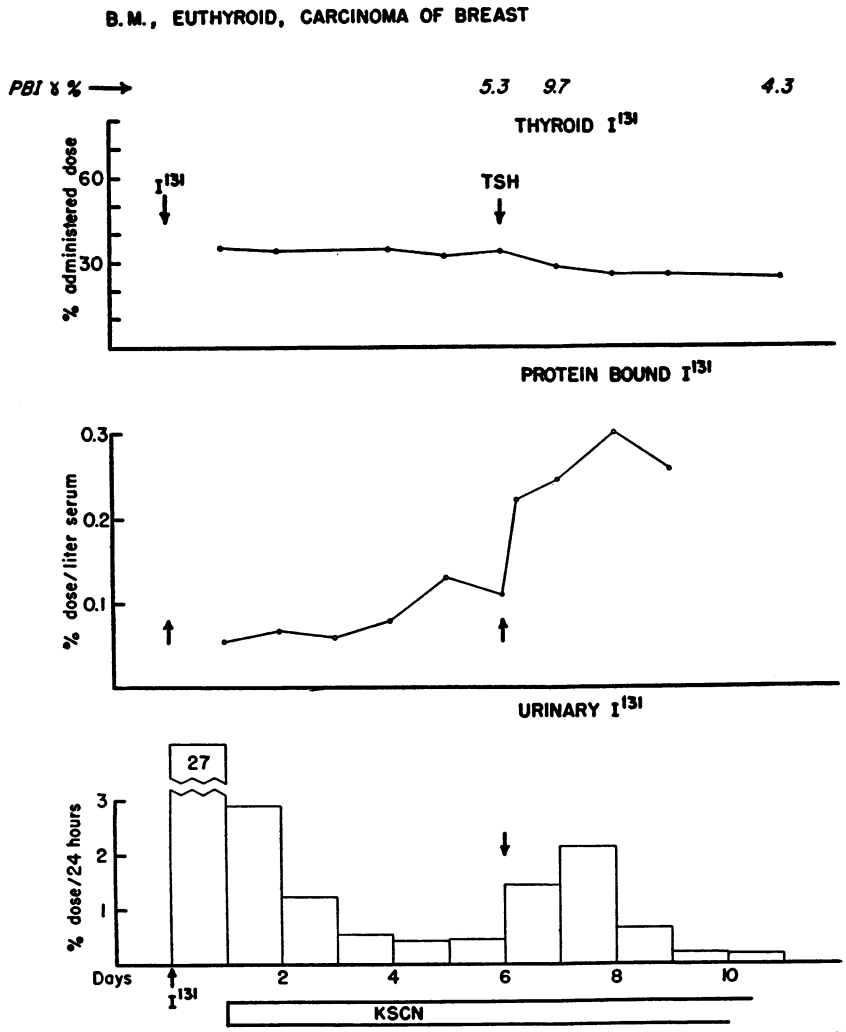

FIG. 2. THE EFFECT OF A SINGLE INJECTION OF TSH ON THE METABOLISM OF I^{131} IN AN EUTHYROID SUBJECT

TABLE I—*Total and precipitable I^{131} in the serum*

Patient		Day after I^{131} administration									Hour after thyroid stimulating hormone administration												
		1	2	3	4	5	6	7	8		2	4	6	8	12	24	48	72	96	120	144	168	
M. D.	[1]T.S. [2]PBI^{131}	0.48 0.036	0.11 0.045	0.074 0.049	0.057 0.044	0.054 0.047	0.055 0.037	0.053 0.043	—* 0.069	Thyroid Stimulating Hormone 40 mg. equiv. I.M.	0.071	0.074 0.045	0.045	0.091 0.071		0.11 0.10	0.11	0.072			0.048		Euthyroid, carcinoma of breast
B. M.	T.S. PBI^{131}	0.46 0.057	0.12 0.067	0.13 0.062	0.14 0.078	0.12 0.13	0.12* 0.11				0.15 0.16	0.20	0.25 0.22	0.30		0.38 0.25	0.30	0.34	0.26	0.20	0.25	0.20	Euthyroid, carcinoma of breast
R. B. (I)	T.S. PBI^{131}	0.35 0.038	0.14 0.061	0.12 0.075	0.12 0.092	0.14*					0.18	0.24	0.30	0.28	0.28 0.23	0.33 0.28	0.31 0.27	0.26 0.24	0.25 0.23	0.22	0.19	0.17	Euthyroid, arteriosclerotic heart disease
J. M.	T.S. PBI^{131}	0.17	0.26 0.25	0.38 0.36	0.40* 0.34						0.40 0.36	0.44 0.39		0.44	0.40	0.42	0.42 0.42	0.40	0.39 0.39				Graves' disease
J. G.	T.S. PBI^{131}	0.21 0.19	0.32 0.29	0.44 0.42	0.51 0.49	0.54* 0.54					0.78 0.70	0.98 0.84	1.05 0.85	1.02 0.84	1.02 0.88	1.14 1.06	0.96 0.83	0.84	0.90 0.82	0.88 0.80	0.91 0.85		Graves' disease
[3]E. G.	T.S. PBI^{131}		0.86 0.83	1.03 0.98	1.09* 1.02							1.37 1.20		1.51 1.35		1.47 1.41	1.25 1.16						Graves' disease
[3]B. V.	T.S. PBI^{131}							0.55 0.40	0.50* 0.52			0.54 0.52		0.56 0.55		0.64							Graves' disease
Y. B. (I)	T.S. PBI^{131}	0.36 0.34	0.51	0.64 0.57	0.60* 0.57						0.80 0.73		0.86 0.77	0.83 0.72	0.77 0.71	0.85 0.74	0.73 0.74	0.63 0.53	0.64 0.57	0.63 0.52	0.60	0.55 0.49	Graves' disease
[4]Y. B. (II)	T.S.	0.39	0.44	0.38	0.49*						0.49	0.57 0.58	0.57 0.57	0.58 0.57		0.62 0.60	0.58	0.58 0.59	0.56 0.52	0.54 0.49			Graves' disease
S. S. (I)	T.S. PBI^{131}	0.16 0.14	0.35 0.28	0.39 0.37	0.37* 0.38						0.46 0.42	0.57 0.51	0.56 0.51	0.56 0.50		0.60 0.59	0.72 0.69	0.69 0.66	0.59 0.55	0.56 0.54		0.44	Graves' disease
[4]S. S. (II)	T.S. PBI^{131}	0.14 0.15	0.22 0.23	0.29 0.30	0.39* 0.37						0.38 0.36	0.42 0.39	0.46 0.43	0.46 0.44		0.49	0.48	0.56 0.56	0.57 0.54	0.71 0.67	0.66 0.64	0.59 0.54	Graves' disease
M. L.	T.S. PBI^{131}	2.57 2.43	3.03 2.83	2.99 2.90	3.03* 2.79							2.86 2.71		3.32 2.98		3.13 2.84	2.82 2.59	2.86 2.56	2.47 2.28	2.66 2.16			Graves' disease
H. D.	T.S. PBI^{131}	1.05 1.06	1.76 1.72	1.83 1.77	2.05* 1.86						2.21 2.00	2.27 1.95	2.29 2.03	2.38 1.92	2.15 1.86	2.23 1.93	2.10 1.91	1.71 1.69	1.50 1.59				Nodular goiter, hyperthyroid (?)
P. O.	T.S. PBI^{131}	2.91 2.84	3.23 3.05	3.06 2.83	2.80* 2.78						2.98 2.82	2.82 2.77		2.82 2.73	3.08 2.89	2.64 2.50	2.45 2.39	2.12	1.90		1.56		Acromegaly, Graves' disease
S. B.	T.S. PBI^{131}	0.11 0.024	0.050	0.061 0.049	0.054 0.036	0.064 0.055	0.066* 0.067					0.11 0.077		0.19 0.089		0.14 0.12	0.13 0.14	0.11 0.10	0.11 0.096				Acromegaly, euthyroid
S. M.	T.S. PBI^{131}		0.16 0.11	0.14 0.093	0.11 0.10	0.14 0.14	0.15*				0.12	0.17	0.19 0.18	0.15 0.15		0.20 0.17	0.21 0.17	0.19	0.17	0.14			Acromegaly, euthyroid
[5]R. B. (II)	T.S. PBI^{131}	0.68 0.040	0.19 0.066	0.14 0.087	0.13 0.11	0.14* 0.11				Cortisone	0.15 0.13	0.13 0.12	0.13 0.12	0.13 0.11	0.15 0.13	0.17 0.13	0.18 0.15	0.16 0.15	0.16 0.15	0.16 0.15	0.16		Euthyroid, arteriosclerotic heart disease

Patient		Day after I^{131} administration												
		1	2	3	4	5	6	7	8	9	10	11	12	
[6]F. F.	T.S. PBI^{131}	0.62 0.034	0.34 0.12	0.23 0.17	0.26 0.21	0.28 0.24	0.31 0.26	0.29 0.26	0.32 0.27	0.33 0.28	0.27	0.26	0.28	Euthyroid, malignant melanoma

All values are in terms of per cent of the administered dose of radioiodine per liter of serum. [1] T.S.—total serum radioactivity. [2] PBI^{131}—protein bound radioiodine.
In all experiments, except those labeled Y. B. (II) and S. S. (II) the patient was given 1.0 gm. of potassium thiocyanate 24 hours after the I^{131} was administered. This medication was continued at a dosage of 0.5 mg. per day for the duration of the experiment.
[3] 10 mg. equivalents of TSH was given in place of the usual 40. [4] Patients Y. B. (II) and S. S. (II) received no thiocyanate.
[5] 500 mg. of cortisone was given orally in place of the usual injection of thyroid stimulating hormone. [6] Neither TSH nor cortisone given.
* The asterisk indicates the value obtained immediately before the administration of the TSH. It thus indicates the day on which the medication was given.

Again, the pituitary preparation produced no effect on the amount of radioactivity measured over the neck. The serum PBI^{131} in this patient showed a more marked response to TSH. An elevation of 130 per cent was produced by the end of 24 hours, and by 48 hours this index had risen over 200 per cent of its "control" value. The PBI^{127} measured at 24 hours showed a rise of 80 per cent. The urinary excretion of I^{131} demonstrated a somewhat different pattern from that seen in the hyperthyroid patient in Figure 1. Here, more of the radioiodine was excreted in the second 24-hour period following TSH, with elevations of 310 and 325 per cent in the first and second 24-hour periods, respectively.

Table I lists all the determinations of the radioactivity in both the total serum and the protein-precipitable fraction of the serum. It can be seen that by 24 hours following the administration of I^{131} almost all the serum radioactivity of the hyperthyroid patients was protein bound. In the euthyroid patients however, the radioactivity did not completely precipitate with the proteins until 4 to 6 days had elapsed after the administration of the I^{131}. Because of this, the euthyroid patients were given TSH between the fifth and eighth day following the tracer dose whereas the hyperthyroid patients received the injection by the fourth day. It can also be noted in this table that the blood levels of radioactivity in the hyperthyroid patients tended to be somewhat higher than those of the euthyroid patients, as was previously noted by McConahey, Keating, and Power (16).

Within two hours after the injection of TSH many of the patients showed an elevation in the blood level of PBI^{131}. A maximum elevation was observed in the hyperthyroid patients within 24 hours after administration of TSH. Euthyroid patients, on the other hand, did not show a maximum response until 48 hours after the administration of TSH. In addition, elevations of the blood levels in the euthyroid patients were somewhat greater than those in the hyperthyroid subjects (Figure 3).

In Table II are listed the measurements of radioactivity in the neck (in terms of per cent of administered dose). With the possible exception of patient J. G., no patient demonstrated any decrease in this index that could be attributed to the administration of the single dose of thyrotrophic material. Although not appearing on this table, measurements were made 2, 4, 6 and 8 hours following the injection and these determinations likewise showed no significant changes.

The biological half-life of the radioiodine in the thyroid (observed half-life corrected for physical decay) ranged between 11 and 50 days with the exception of 3 patients (Table II). These patients (P. O., H. D., and M. L.), the only subjects showing insignificant response to the injected TSH, had an extremely rapid half-life of I^{131} in the thyroid (5.0, 2.5 and 6.3 days, respectively).

In Table III are shown values for the urinary excretion of radioiodine per 24-hour period in terms of percentage of the dose administered. These determinations fall into the usual patterns for euthyroid and hyperthyroid patients. Following administration of the TSH preparation, a considerable elevation in the urinary radioactivity was demonstrated in all except 3 of the patients studied. These patients (P. O., H. D., and M. L.), previously noted for their high turnover rates of

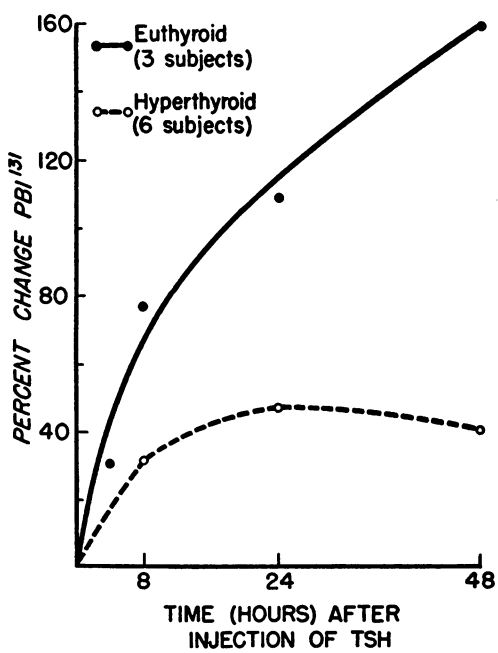

FIG. 3. THE EFFECT OF TSH ON THE LEVEL OF THE SERUM PBI^{131}

Values are calculated in terms of per cent increase in PBI^{131} as compared to a control value taken immediately before the injection of TSH.

TABLE II

Radioactivity measured over the neck

Patient	Dose I^{131}	Day after I^{131}									Day after TSH					Biological half-life of I^{131} in thyroid
		1	2	3	4	5	6	7	8		1	2	3	4	5	
	μc															*days*
Euthyroid																
M. D.	.594	16.4	21.1	20.4	19.3	19.8	17.8	20.0	15.9*		15.1	15.9	16.0	14.2		22
B. M.	.685	35.0	33.9		33.8	32.6	33.0*				28.2	25.1	24.9		23.7	18
R. B. (I)	.740	30.6	29.0	34.6	31.6	27.8*					24.6	25.8		25.0	25.2	31
Graves' Disease																
J. M.	.476	64.3		72.5	54.2*						53.3	61.7	58.6			20
J. G.	.917	68.6	74.7	72.4	68.1	73.4*					55.4	52.2	59.0		56.4	23
[3]E. G.	.500		72.5													
[3]B. V.	.600		65.2													
Y. B. (I)	.528	70.0	56.8	73.9	66.0*						57.1		58.0	52.1	53.9	38
[4]Y. B. (II)	.870	68.5	61.9		49.5*						48.0	43.7	51.2	56.0	53.3	16
S. S. (I)	.456	40.4	47.7		42.7*						35.7	33.2	28.9	30.0	28.5	11
S. S. (II)	.815	55.9	47.7		50.6*						39.6	43.3	42.5	42.0	39.7	16
M. L.	.100	49.2	43.0	37.3	34.9*						30.5	28.6		27.6	23.1	6.3
H. D.	.800	54.4	37.3	29.6	25.0*						15.5	11.3	10.0			2.5
Acromegaly																
P. O.	.820	44.7	30.8	24.1	19.9*						15.2	15.3	13.4	10.2		5.0
S. B.	.497	32.4	30.3	30.2	31.9		27.4				25.0	25.0	25.6	25.3		20
S. M.	.502	26.6	20.3	19.7	20.0	19.0*					17.1	14.9	16.5	16.7		27
[5]R. B. (II)	.950	20.7	22.0	22.6	22.8	22.6*				Cortisone	21.1	20.7	20.7	19.0	19.5	50

Control		Day 1	2	3	4	5	6	7	8	9	10
[6]F. F.	.644	32.4	32.5		26.6		26.0	26.4	26.3	24.0	24.0

All values are in terms of per cent of administered dose of radioiodine.
All other notations are the same as those on Table I.

radioiodine, will be considered as a separate group and are excluded from the general discussion of the effects of TSH. Their data are, consequently, not included in Figures 3 and 4. In terms of percentage increase over "control" values for the various indices, the effect of TSH was the greatest in the urine radioactivity measurements. Here, increments ranged from 100 to 350 per cent as compared to elevations in the serum PBI^{131} which ranged from 30 to 200 per cent. Again, it may be noted that the hyperthyroid patients reached their maximum increments within the first 24 hours following thyrotrophin injection while the euthyroid patients often excreted considerably more radioactivity in the second 24 hours. Figure 4 shows the changes in the average percentage increment in urinary I^{131} in the three 24-hour periods following injection of TSH.

Table IV shows the values for the serum protein-bound iodine determined chemically. These values show elevations of varying degrees following the administration of TSH. From these data and the PBI^{131}, it is possible to calculate the specific activity (the ratio of atoms of radioactive isotope to atoms of stable isotope) of the iodine of the blood before and after the administration of the thyrotrophin. These figures (Table V) remain relatively constant for each patient.

DISCUSSION

TSH has been previously shown to cause the secretion of thyroid hormone by the thyroid gland in animals and in patients both normal and hyperthyroid (3, 6, 8–10, 19). The present study differs from previously reported investigations in attempting to quantitate the variations in response to a standard dose of TSH given to normal, hyperthyroid and acromegalic man.

TABLE III

Urinary excretion of radioiodine

Patient	Day after I^{131}									Day after TSH						
	1	2	3	4	5	6	7	8		1	2	3	4	5	6	7
Euthyroid																
M. D.	62.0	9.89	1.65	.716	.515	.382	.291	.303*		1.25	1.28	.71	.421	.333	.252	.204
B. M.	26.9	2.90	1.22	.559	.442	.450*				1.47	2.13	.875	.210	.205	.178	.168
R. B. (I)	43.5	6.24	1.85	.900	.660*					2.08	2.88	1.02	.350	.340	.360	.320
Graves' Disease																
J. M.	5.18	.559	.895	.709*						1.69	.891	.825	.737			
J. G.	9.36	.574	.803	.952	1.04*					4.51	2.23	.924	.900	1.00	1.05	
[3]E. G.	10.5	.403	.386	.962*						2.58	1.85					
[3]B. V.	22.4	.790						.340*		.953	.360					
Y. B. (I)	12.8	.791	.802	1.04*						2.72	1.49	1.18	.905	.995	.700	.631
[4]Y. B. (II)	30.4	1.57	1.03	1.06*						2.95	1.95	1.11	1.11	1.67	1.20	
S. S. (I)	45.7	1.25	1.34	1.55*						2.66	(2.00)	2.18	1.26	1.06	.800	
[5]S. S. (II)	46.4	.918	.622	.645*						.995	.625	.591	.635	.385	.580	
M. L.	12.6	3.01	2.42	3.31*						4.35	2.32	2.26	1.74	1.23		
H. D.	24.8	5.95	5.90	4.70*						7.12	4.80	3.12	2.86	2.49	2.42	
Acromegaly																
P. O.	16.8	3.09	5.15	4.03*						3.95	4.04	3.57	3.16	3.29	2.44	2.43
S. B.	33.5	2.41	.630	.420	.360	.374*				1.33	.591	.185	.116			
S. M.	57.8	7.75	1.73	.360	.617	.526*				1.40	.990	.572	.273	.291		
[6]R. B. (II)	56.9	11.9	1.66	.851	.577*				Cortisone	.508	.390	.431	.378	.393	.412	

Control	Day 1	2	3	4	5	6	7	8	9	10
[6]F. F.	41.1	7.99	2.74	1.01	.910	.994	.973	.951	.949	.910

All values are in terms of per cent of administered dose per 24 hour period.
All other notations are the same as those on Table I.

TABLE IV

Protein-bound iodine (PBI^{127}) in serum
(All values in γ%)

Patient	Control immediately before TSH	8-hours post-TSH	24-hours post-TSH	48-hours post-TSH	72-hours post-TSH
Euthyroid					
M. D.	5.4	6.9		9.6	
B. M.	5.3		9.7		
R. B. (I)	8.7	15.3	18.4		9.5
*R. B. (II)	8.3	5.2	3.3	5.8	
Graves' disease					
J. M.	17.8	18.9		19.8	
J. G.	11.4	15.0			
E. G.	15.3		18.7		
B. V.	20.5		24.6		
Y. B. (I)	12.2	16.1			
S. S. (I)	4.7	6.1		9.5	
H. D.	6.3	6.5		6.9	
M. L.	16.6	20.1	19.1	16.1	
Acromegaly					
P. O.	5.5	4.1			
S. B.	5.6		9.2		
S. M.	9.5	10.6	12.7	14.2	11.3
Control	Day 6	Day 9	Day 10		
F. F.	3.5	3.0	2.8		

* This patient received cortisone (500 mg.) instead of TSH.

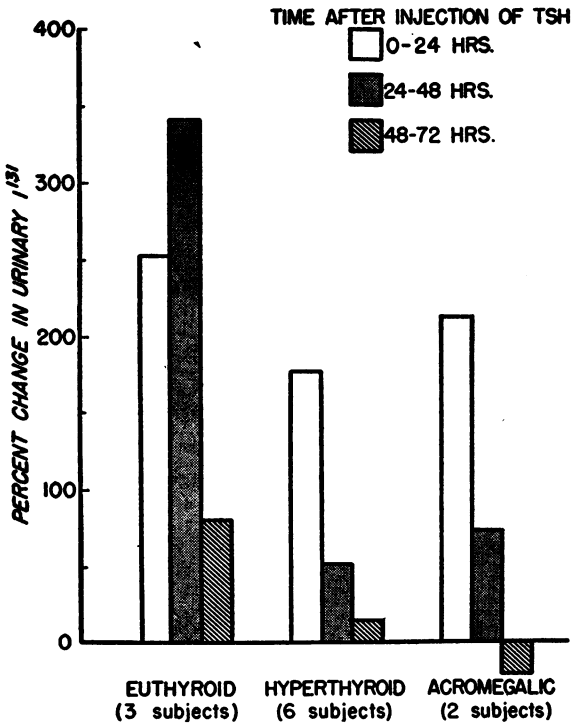

FIG. 4. THE EFFECT OF TSH ON URINARY EXCRETION OF RADIOIODINE

Values are calculated in terms of per cent change in 24-hour urinary excretion as compared to the "control" 24-hour period immediately prior to the administration of the TSH.

The negative value seen on the third day in the acromegalic patients represents an average value below that obtained on the control day.

Certain difficulties are inherent in the use of TSH at the present time and in the quantitation of this study. The preparation of TSH used was obviously impure and may have been contaminated with small amounts of other pituitary hormones. Since there was no change in the volume of urine secreted before and after the administration of TSH, it appears unlikely that antidiuretic hormone was present in sufficient amount to interfere with the study. Eosinophile counts were performed before and 4 hours after the TSH to evaluate the possibility that ACTH was a major contaminant. In three of five patients studied there was no significant change in eosinophile count after TSH but in two patients there was a significant fall. A further control was done to see if adrenocortical hormones whose secretion might have been caused by ACTH would affect the secretion of thyroid hormone. In one patient (R. B.–II) 0.5 gram of cortisone was administered orally at the time TSH was usually given. No change was seen subsequent to this in the serum, urinary, or thyroidal radioiodine. Another possibility is that TSH (or contaminants) might affect the rate of breakdown, excretion, or distribution of thyroid hormone. In one patient TSH was administered 10 days after intravenous injection of thyroxine labeled with I^{131}. There was no change in the level of radioactivity in the blood or urine that could be attributed to TSH.

Thiocyanate might have a specific effect on the response of the thyroid to TSH. In two hyperthyroid patients (Y. B.–II and S. S.–II) the usual experimental protocol was repeated except that the administration of potassium thiocyanate was omitted. Only random deviations were noted in these experiments indicating that the thiocyanate had no specific effect on the response of the gland to thyroid-stimulating hormone. Since iodide released upon de-iodination of the thyroid hormone is in part accumulated by the thyroid, one would expect a greater urinary excretion of iodine if thyroidal reaccumulation is prevented (as by thiocyanate). This occurred with S. S. but not with Y. B. (Table III).

TABLE V

Iodine in thyroid and iodine discharged by TSH

Patient	Iodine discharged subsequent to TSH	Estimated I^{127} in thyroid	Thyroidal iodine discharged by TSH
	mg.	*mg.*	%
Euthyroid			
M. D.	2.27	12.4	18.3
B. M.	2.95	15.9	18.6
R. B.	4.31	22.0	19.6
Graves' disease			
J. M.	1.17	28.4	4.12
J. G.	1.60	15.7	10.2
E. G.	0.544	—	—
Y. B. (I)	0.881	14.1	6.18
S. S. (I)	0.804	5.3	15.1
*H. D.	—	0.85	—
*M. L.	—	2.1	—
Acromegaly			
*P. O.	—	0.39	—
S. B.	1.81	22.9	7.91
S. M.	1.09	12.9	8.45
Control			
F. F.	—	2.5	—

* Patients H. D., M. L., and P. O. did not respond to administered TSH.

Another possible source of error is failure of thiocyanate to inhibit iodine uptake by the thyroid. The interpretation of the data is contingent upon the assumption that there is no recirculation of iodine from the catabolized thyroid hormone back into the thyroid. Thiocyanate levels were usually maintained at 4 to 5 milligrams per cent. However, somewhat lower levels of thiocyanate appeared to be partially effective. In one patient (B. M.) there was 35 per cent uptake of a tracer dose of I^{131} which was depressed to 7 per cent uptake where the level of thiocyanate was 1.8 milligrams per cent. Another tracer showed 4.8 per cent uptake when the level of thiocyanate in the blood was 3.1 milligrams per cent. It should be noted that even in an athyreotic patient the radioiodine in the tissues of the neck gives an apparent uptake of about 2 per cent. If the uptake of iodine is not inhibited, the level of radioiodine in the precipitable fraction of the serum rises, and by 4 to 10 days falls again. If, on the other hand, recirculation of iodine is prevented, the level of precipitable I^{131} in the serum will increase asymptotically and not fall until the entire store of iodine in the thyroid is exhausted. In control studies (F. F., R. B.–II) it is apparent that the level of PBI^{131} reaches a plateau and does not fall. This is fairly conclusive evidence that the recirculation of iodine was prevented by the thiocyanate.

If the recirculation of radioiodine has been prevented by thiocyanate then the specific activity of the precursor, which in this case is thyroglobulin stored in the thyroid, is maintained constant. Zilversmit has described mathematically the sequence of events in the two situations when the specific activity (S. A.) of the precursor varies and when it is constant (20). In the first case the specific activities of the precursor and product are identical at only one point, the maximum S.A. of the product. In the second case, however, the S.A. of the product approaches asymptotically the S. A. of the precursor. As can be seen in the two control studies (F. F. and R. B.–II) the rate of increase in the S.A. of the product is quite rapid so that by 4 to 7 days after administration of the tracer a plateau is reached for all practical purposes. At any time thereafter it is possible to calculate iodine in the thyroid since the S.A. of the product and the activity of the precursor are known. These data are shown in Table V. If any iodine in the thyroid is in a metabolically inactive pool and therefore is not labeled by the I^{131}, the values calculated for thyroidal iodine will be too small by a factor related to the size of the inert iodine pool.

In general it appears that euthyroid patients have somewhat more iodine stored in their thyroid than do hyperthyroid patients. Patient J. M. had been treated with Lugol's solution intermittently for 10 years prior to the present study and it is not surprising to find, therefore, that there appeared to be a larger amount of iodine in his thyroid. The average values for both euthyroid and hyperthyroid patients are somewhat greater than those found on actual analysis of total iodine content of thyroids as determined by Gutman, Benedict, Baxter and Palmer, who found an average of 8.8 milligrams in normal thyroids and 5.1 milligrams in thyroids of hyperthyroid patients who had not been treated with iodine (21). No explanation is immediately apparent to explain this discrepancy.

From the time of plateau of the serum PBI^{131} until exhaustion of iodine stores in the thyroid a steady state prevails with respect to both the I^{127} and I^{131} in the serum. One would expect, therefore, that the specific activity of the serum would remain relatively constant. That this did occur is seen in Table VI and a measure of the constancy in S.A. for each patient is the fact that the average deviation of these values from their own mean is 9 per cent. Since the steady state lasts for at least a week it is possible to calculate the magnitude of an increment in serum PBI^{127} and PBI^{131}, if this is of relatively short duration. For calculation of the total increment, however, the volume of distribution of the PBI^{131} at the time of calculation must be known. Albert and Keating have shown that by 48 hours after intravenous administration of thyroxine labeled with I^{131} the early rapid disappearance into the volumes in which it is distributed is essentially complete. At this time the volume of distribution is approximately 20 per cent of the body weight (22). This figure can be used in calculating the volume of distribution of the PBI^{131} 48 hours after TSH. A certain fraction of the hormone discharged by TSH will, in 48 hours, have been excreted in the urine. Since the serum is the only source of urinary iodine the S.A. of serum and urine are as-

TABLE VI

Specific activity of protein-bound iodine in serum
$\mu c/\gamma$

Patient	Control	Hours after TSH injection 8	24	48	72	96	144
Euthyroid							
M. D.	0.0076	0.0061		0.0086			
B. M.	0.014	0.030	0.018				
R. B. (I)	0.0094	0.012	0.011				
*R. B. (II)		0.020	0.023	0.024			
Graves' disease							
J. M.	0.0091			0.010			
J. G.	0.043	0.0513					
E. G.	0.033		0.038				
B. V.	0.015		0.018				
Y. B.	0.025	0.024				0.021	
S. S.	0.037	0.037		0.033			
H. D.	0.25	0.23	0.28				0.29
M. L.	0.016	0.015	0.016	0.018			
Acromegaly							
P. O.	0.41	0.55					
S. B.	0.0060		0.0065				
S. M.	0.0079	0.0071	0.0079	0.0074	0.0084		

Control	Days after tracer 9	10	14
F. F.	0.060	0.062	0.064

* This patient received cortisone instead of TSH.

sumed to be identical. Then the following equation gives an approximate value for the iodine discharged by the single injection of TSH (over and above that which was normally secreted).

$$I^{127} = \left(\frac{\Delta PBI^{131} \times cPBI^{127}}{cPBI^{131}}\right) \times .2\ BW + \left(\frac{\Delta UI^{131} \times cPBI^{127}}{cPBI^{131}}\right)$$

where I^{127} = iodine discharged during the 2 days subsequent to TSH. This does not include the iodine secreted at the previously established rate for that patient at that time,

ΔPBI^{131} = increment in PBI^{131} of serum 48 hours after TSH in per cent of dose per liter of serum,

$cPBI^{127}$ = PBI^{127} in serum prior to TSH in micrograms per liter of serum,

$cPBI^{131}$ = PBI^{131} in serum immediately before the injection of TSH in per cent per liter of serum,

BW = body weight in kg.,

ΔUI^{131} = total increase in I^{131} in the urine during the 48 hours after the TSH in per cent of dose.

The amount of iodine discharged from the thyroid in response to a single injection of TSH as calculated above is shown in Table V. It is apparent that the euthyroid patients secreted more iodine in response to the same amount of TSH than did the hyperthyroid patients. The patients with acromegaly were intermediate between these two groups. It is of some interest, furthermore, that the hyperthyroid patients secreted a smaller proportion of iodine stored in their thyroid than did the euthyroid patients. If the patients with hyperthyroidism had increased levels of circulating TSH it would appear reasonable for a standard dose of TSH to produce less effect in these patients than in those who had a normal level of circulating TSH. Certainly such an explanation is little more than speculation without more direct data.

An additional feature of interest in which hyperthyroid and euthyroid patients differed was in the rapidity of response to TSH. As can be seen in Figures 3 and 4 the effect of TSH was not maximal in euthyroid patients until 48 hours after its administration, whereas in hyperthyroid cases the maximum effect was reached in 24 hours.

There were three (P. O., H. D. and M. L.) who showed no response to TSH. These patients, however, had such a small amount of iodine stored in the thyroid that were 10 per cent of it secreted in response to TSH (the average percentage of thyroidal iodine released from this

dose of TSH was 9 per cent for hyperthyroid patients) the increment in urinary or serum iodine would have been very difficult to detect. One cannot therefore consider these patients to have been unequivocally non-responsive to TSH. However, the pattern of iodine metabolism in these cases was quantitatively markedly different from that in the other patients. All three had very low thyroidal iodine, an unusually fast disappearance rate of iodine from the thyroid (6.3, 2.5 and 5.0 days) and abnormally elevated levels of PBI^{131} in the serum (2 to 3 per cent of dose per liter). These cases might represent a basically different physiology from the other cases of Graves' disease studied.

The patients with hyperthyroidism had varying degrees of exophthalmos from J. B. and H. D. who had none, to S. S. whose exophthalmos was very severe and who had recurrent corneal ulcerations. There appeared to be no correlation between the degree of exophthalmos and any of the parameters of iodine metabolism studied.

There is no clear explanation for the relatively greater increase in urinary I^{131} after TSH than in serum PBI^{131} or total I^{131}. At least two possibilites arise: *1*) Thyroid hormone and its iodine-containing metabolites are cleared by the kidney at rates dependent on their concentration in serum, or *2*) after TSH there is a relative increase in concentration in the serum of an iodine-containing compound which is rapidly cleared by the kidney. There are no data available bearing on this matter.

CONCLUSIONS

1. The effect of a single intramuscular injection of thyroid stimulating hormone on the metabolism of a tracer dose of I^{131} was studied in three euthyroid, eight hyperthyroid and three acromegalic patients.

2. When administered four to eight days after the tracer of I^{131}, TSH increased the level of PBI^{131} and PBI^{127} in the blood and I^{131} in the urine.

3. Methods are presented for the estimation of the total amount of thyroid hormone discharged from the thyroid after the injection of TSH and for calculation of the total iodine content of the thyroid.

4. Following a standard dose of TSH, euthyroid patients appeared to secrete more thyroid hormone than did hyperthyroid patients. The maximum effect of TSH appeared in the first 24 hours in the hyperthyroid patients, and not until 48 hours in euthyroid patients.

5. Three hyperthyroid patients are presented who demonstrated an unusual method of handling radioiodine. This is characterized by a very rapid turnover rate of iodine by the thyroid with consequent abnormally elevated serum protein-bound radioiodine and urinary radioiodine levels.

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