CLINICAL AND PHYSIOLOGICAL OBSERVATIONS IN A PATIENT WITH AN IDIOPATHIC DECREASE IN THE THYROXINE-BINDING GLOBULIN OF PLASMA *

By SIDNEY H. INGBAR †

(From the Thordike Memorial Laboratory, Second and Fourth (Harvard) Medical Services, Boston City Hospital and the Department of Medicine, Harvard Medical School, Boston, Mass.)

(Submitted for publication June 13, 1961; accepted August 4, 1961)

Since its discovery in 1952 (1), the thyroxine-binding globulin of plasma (TBG) has been the subject of extensive investigation designed to elucidate its physicochemical characteristics, alterations in abnormal states, and physiological function. Recent reviews have summarized available knowledge in this area (2, 3).

The present report describes a male patient whose plasma appears to be virtually devoid of thyroxine binding by TBG. This unusual abnormality, fortuitously discovered, afforded a unique opportunity to assess further the role of TBG in regulating the peripheral metabolism of the thyroid hormone. A number of physiological studies was therefore performed in this patient, and the results are reported herein. While these studies were in progress, clinical features of a male patient with a similar thyroxine-binding abnormality were reported by Tanaka and Starr (4). It is likely that the physiological disturbances currently described were also present in the earlier case and that both patients are affected by the same distinct, though rare, disorder in plasma protein metabolism.

CASE HISTORY

History. F.B., a 58 year old unmarried white male, was admitted to the Harvard Medical Service of the Boston City Hospital on September 14, 1958. His chief complaint at that time was increasingly severe exertional dyspnea, orthopnea, and episodes of paroxysmal nocturnal dyspnea of 2 years' duration. He had taken digitalis irre-\[...\]

* This investigation was supported in part by Research Grant A-267 from the National Institute of Arthritis and Metabolic Diseases, and in part by the Medical Research and Development Board, Office of The Surgeon General, Department of the Army, under Contract DA-49-007-MD-412. Read by title at the Annual Meeting of the American Society for Clinical Investigation, May 2, 1960, Atlantic City, N. J.

† Investigator, Howard Hughes Medical Institute.
nuchoid. There was a slight temporal calvities, but no other balding.

There were no peripheral stig mata of hyper- or hypothyroidism. There were no pigmentary abnormalities. The eyes were highly myopic, but there were no cata
racts. Visual fields were normal to gross confrontation.

There was moderate weakness of the distal musculature of the arms and legs with atrophy of the intersosseous muscles. No myotonia was evident. Deep tendon reflexes and sensory examination were normal.

Laboratory. The urine and blood were within normal limits. Lumbar puncture was normal. Serum chemis-
tries included: urea nitrogen, 15 mg per 100 ml; glucose (fasting), 100 mg per 100 ml; calcium, 10.2 mg per 100 ml; phosphorus, 3.1 mg per 100 ml; cholesterol, 210 mg per 100 ml; sodium, 138 mEq per L; potassium, 4.1 mEq per L; chloride, 104 mEq per L; CO₂, 25.6 mEq per L. Serum iron concentration was 110 μg per 100 ml and iron-binding capacity 320 μg per 100 ml, both within normal limits.

Skull films were normal. A bone survey revealed no abnormalities; the epiphyses of the iliac crests were closed. Visual fields were within normal limits. Twenty-

four hour urinary excretion of 17-ketosteroids was 9.9 mg (5), of 17-hydroxycorticoids, 4.3 mg (6). Urinary gonadotropins were positive at 50 and equivocal at 100 mouse units per 24 hours (7).

Twenty-four hour thyroidal uptake of I²⁺ was 28 per cent. BMR's ranged between −5 and +4 per cent. Serum protein-bound iodine (PBI) was 2.0 μg per 100 ml (8).

Course. The patient's cardiac complaints responded well to digitalization and a low salt diet. Mercurial di-
uretics were not employed. Cardiac catheterization studies were interpreted as consistent with a persistent left
vena cava and atrial septal defect, probably of the secundum type.

In view of the patient's obvious hypogonadism, a thor-
ough endocrine evaluation was undertaken. The associated subnormal PBI, repeatedly confirmed, strongly raised the possibility of pituitary hypofunction, but this diagnosis seemed clearly excluded by the neurological and X-ray examinations and by other endocrinological findings, especially the increased urinary excretion of gonadotropins. The dissociation between the PBI and other laboratory, as well as clinical, indices of thyroid function raised the possibility of abnormal binding of thyroxine in the patient's plasma. This hypothesis was confirmed by repeated electrophoretic studies of the pa-
tient's serum, all of which revealed a virtual absence of TBG.²

Studies of the peripheral turnover of I²⁺-labeled thy-
roxine were then performed. Several days after these were complete, the left inguinal hernia became incarcer-
ated and emergency repair was performed. At this time the testis was seen to be grossly atrophic and a testicular biopsy was taken. In view of the muscular weakness, a biopsy of the anterior tibial muscle and overlying skin was also made. The latter revealed no abnormality. Histologically, the testis was fibrotic, with atrophy and fibrous replacement of tubular elements and decrease in interstitial cells, consistent with chronic restriction of blood supply. Genetic sex was masculine, as assessed in both testicular and cutaneous biopsies.

When the patient's convalescence was complete, oral administration of diethylstilbestrol, 30 mg daily, was begun. This was continued for 7 weeks. During the lat-
ter portion of this period, the patient developed increasing dyspnea, pretibial edema, marked areolar pigmen-
tation, and pronounced tender gynecomastia. Urinary ex-
cretion of gonadotropin declined to less than 5 mouse U per 24 hours. Electrophoretic studies of hormonal bind-
ing in the patient's serum were performed at weekly in-
tervals during the administration of diethylstilbestrol. During the last 10 days of this regimen, studies of the peripheral metabolism of labeled thyroxine were repeated.

Two months later, after clinical evidence of estrogenic effect had disappeared, a course of adrenocortical sup-
pressive therapy was begun. The patient was given 10.0 mg prednisolone ³ by mouth, each day for a period of 6 weeks. Under this treatment urinary 17-ketosteroids de-
clined to 1.1 mg daily. Bloods were drawn weekly for elec-
trophoretic analysis of thyroxine binding.

After gradual withdrawal of corticoid therapy, a 6-week course of sodium-L-thyroxine, 0.2 mg by mouth daily, was instituted. The patient experienced no symptoms from this medication; physical examination, PBI, and electrophoretic findings with regard to thyroxine binding remained unaltered. Twenty-four hour thyroidal uptake of I²⁺ was suppressed from its control value of 28 per cent to 6 per cent, however.

After withdrawal of exogenous thyroid hormone, the pa-
tient remained well and has continued thus, save for one episode of congestive heart failure that followed dis-
continuance of digitals, which quickly responded to re-
digitalization. Bloods have subsequently been obtained at intervals for electrophoretic analysis; these have con-
tinued to show a virtual absence of TBG.

MATERIALS AND METHODS

Thyroxine-binding capacities of individual serum pro-
teins were determined by zonal electrophoresis in filter paper of serum enriched with variable quantities of I²⁺-labeled and stable L-thyroxine.

referred to as a change in TBG; no implications con-
cerning the absolute concentration of the protein are intended.

³11β, 17α, 21-trihydroxy-4-pregna-3, 20-dione.
1³°-labeled hormones (l-thyroxine, T₄; 3,5,3'-l-triiodothyronine, T₃; 3,5,3',5'-triiodothyroacetic acid, triac; 3,5,3',5'-tetraiodothyroacetic acid, tetrac) obtained from a commercial source,⁵ were 90 to 95 per cent pure, as assessed chromatographically, and were diluted with 1.0 per cent human serum albumin to a final concentration of 160 μc per ml immediately upon arrival. This procedure reduced both the degradation of labeled compounds and their adsorption to glassware. Nevertheless, labeled materials were not employed for studies after 1 week of storage in this form.

The procedure employed in preparing samples of serum for the electrophoretic estimation of thyroxine binding by TBG and thyroxine-binding prealbumin (TBPA) was as follows:

To 6.2 ml of serum, 75 μl of 1³°-labeled T₃ in human serum albumin was added, yielding a final concentration of approximately 2 μc per ml (Solution I). A 30-ml aliquot of Solution I was pipetted into a separate tube, and to this was added 100 μl of solution of 1.0 per cent human serum albumin containing 14.0 μg of stable T₄. This yielded a solution containing 451 μg of added stable T₄ per 100 ml (Solution G). Aliquots of Solution I, Solution G, and mixtures of the two were employed to prepare samples containing 87, 112.5, 130, 340, 395, and 451 μg of added stable T₄ per 100 ml. The three lower concentrations were employed to calculate the binding capacity of TBG and the three higher concentrations, the binding capacity of TBPA.⁶ Two additional samples of serum containing 1³°-labeled but no stable T₄ were prepared. The first, Solution A, is a mixture of equal parts of Solution I with fresh serum (1 μc 1³° per ml) and the second (Solution H) a 1:10 dilution of Solution A with fresh serum (0.1 μc 1³° per ml). In general, the specific activity of the 1³°-labeled T₃ is such that these two samples contain approximately 4 μg and 0.4 μg of added stable T₄ per 100 ml, respectively. Electrophoresis was performed in sheets of Whatman no. 3 filter paper, 22.5 × 12 inches, large enough to accommodate the 8 separate specimens. After being moistened in buffer medium and blotted, filter paper sheets were placed upon a carefully ground, siliconized glass plate,¹ 15 inches long, 13 inches wide, and 1 inch thick. Aliquots of serum (20 μl) were then applied in thin lines, equidistant from the ends of the sheet, to areas carefully predetermined from a template. Usually, duplicate aliquots of serum were applied to a second filter-paper sheet carefully placed directly upon the first sheet. A minute quantity of bromphenol blue (BPB) was applied to each serum band to serve as a marker for the electrophoretic migration of albumin. Sheets were then covered by a second glass plate, similar in dimension to the first. When, as is often the case, it is desired to analyze an additional 8 samples upon the same machine, a thin glass plate (13 × 15 × ¼ inch) is placed upon the first pair of duplicate sheets. A second pair of sheets containing the additional samples can then be placed upon the thin plate and the entire "sandwich" covered by the second thick glass plate. To minimize evaporation, the hiatus between the top and bottom glass plates at the front and rear of the apparatus was sealed with cellophane tape. At the lateral margins of the plates, filter paper sheets largely filled this hiatus and were allowed to hang into buffer baths. Gravitational flow of buffer across the filter paper was eliminated by means of a horizontal leveling device in the buffer reservoirs.

Buffer systems employed in the present studies were either 0.05 M veronal or a (hydroxymethyl) ammonium (Tris) maleate buffer, both at pH 8.6.⁸ Through platinum electrodes immersed in the buffer reservoirs, a potential of 110 was applied across the filter paper for a period sufficient to produce approximately a 4-inch migration of the serum albumin. This usually required about 18 hours at normal room temperature. No attempts were made to cool the apparatus, as heating is only slight under these conditions.

When migration of proteins was sufficient, filter paper sheets were dried. One sheet was set aside for radioautography and subsequent staining with BPB, making possible the localization of major protein components. The second sheet was cut into 8 strips and, except in the case of Sample H, radioactive scanning of individual electrophoretograms was performed by means of a continuous scanner and dual-channel recorder. Areas under the individual peaks were determined with an electronic integrator whose output is recorded synchronously with the scan itself. The distribution of labeled T₄ was calculated from the recorded integral as the proportion of added hormone bound by each of the demonstrated binding proteins. The total quantity of labeled T₄ bound by each protein per unit volume of serum was calculated as the product of the proportion bound and the estimated total concentration of T₄ in the specimen. The latter estimate is derived from the total concentration of added T₄ (1³°-labeled plus stable) plus an estimate of endogenous T₄, assuming the endogenous PB1 to be comprised entirely of this hormone. Sample H was divided into segments 0.5 cm wide; these were counted individually in a well-type scintillation counter.

In view of the decreased thyroxine-binding capacity of TBG in the present patient's serum, samples were also assayed which had been enriched with both labeled and stable T₄ in a number of concentrations below the usual lowest enrichment with 87 μg of stable T₄ per 100 ml.

The concentration of the major electrophoretically distinguishable groups of plasma proteins was assessed by densitometric scanning of electrophoretic strips stained with BPB.

⁵ Abbott Laboratories, Oak Ridge, Tenn.

⁶ Evidence that thyroxine-binding sites of TBG and TBPA in normal serum are saturated at the concentrations indicated has been presented briefly in a previous report (2) and will be presented in detail in a later communication.

¹ Available from the Pittsburgh Plate Glass Co., Boston, Mass.
Thyroxine turnover studies. Analyses of the peripheral turnover of I\textsuperscript{131} labeled T\textsubscript{4} in vivo were performed according to methods described in detail elsewhere (9). Serial samples of serum and complete 24-hour urinary collections were obtained daily for 8 to 10 days after the intravenous administration of a tracer dose of I\textsuperscript{131} labeled T\textsubscript{4}. These were analyzed for their concentration of I\textsuperscript{131}. Aliquots of urine were also subjected to a butanol extraction procedure, similar to that described for serum (10), and the proportion of total urinary I\textsuperscript{131} present as butanol-extractable I\textsuperscript{131} was determined. The several functions calculated and their derivations are described in the Appendix.

Red blood cell uptakes of I\textsuperscript{131}-labeled hormones. Blood was drawn from a single normal donor (Type O) into a heparinized syringe. After centrifugation, plasma was removed and the erythrocytes were washed 4 times in 3 volumes of normal saline. After the final wash, sufficient normal saline was added to bring the red blood cell suspension to a standard hematocrit. A sufficient volume of the suspension was pipetted into a series of 13 x 100 mm counting tubes to provide 0.5 ml of packed red blood cells in each. Samples of fresh plasma (oxalate anticoagulant) from the patient and from a group of normal control subjects were enriched with 0.62 μCi per ml of I\textsuperscript{131}-labeled compounds; 0.5-ml aliquots of plasma were pipetted into tubes containing the packed red blood cells. Tubes were stoppered and agitated at 37° C for 1 hour. The radioactivity in each tube was then assessed directly in a well-type scintillation counter. Tubes were centrifuged, the plasma removed by suction and the red blood cells were then washed 5 times with 3 volumes of physiological saline solution at room temperature. After the last wash, 0.5 ml of saline was added to the packed red blood cells to bring the final volume to 1 ml. The cells were resuspended, and the radioactivity in the tube was again measured.

Red blood cell uptakes of labeled hormones were calculated as the ratio of final to initial counting rates, expressed as a per cent. All analyses were performed in duplicate. Electrophoretic analyses of thyroxine-protein-binding interactions in the sera studied were performed in all instances.

RESULTS

Electrophoretic analyses (Figures 1, 2; Table 1). Repeated electrophoretic analyses of the patient's serum failed to reveal more than a trace of T\textsubscript{4} in the inter-α-globulin (TBG) area, during electrophoresis of serum in either the Tris-maleate or the veronal buffer systems.\textsuperscript{9} Radioactivity in the TBG zone was most frequently found when electrophoretograms of serum enriched with only 0.4 μg of T\textsubscript{4} per 100 ml were cut into strips and counted in a well counter. Even at this concentration, however, no more than an approximate 5 per cent of total radioactivity was localized in the TBG zone. Often, both before and during administration of diethylstilbestrol, no TBG could be demonstrated. In sera enriched with approximately 4.0 μg of T\textsubscript{4} per 100 ml, a small upward convexity in the radioactive scan was occasionally seen in the TBG zone. This was evident on radioautographs as a faint zone of darkening. Direct well counting of such strips indicated that no more than 2 to 3 per cent of added radioactivity was localized with TBG. In sera enriched with higher concentrations of T\textsubscript{4}, no band in the TBG zone could be discovered. It thus appears likely that this patient's serum was either not completely or not always devoid of TBG. The binding capacity of TBG, when demonstrable, could not be assessed with any accuracy but clearly seemed no more than a small fraction of 1 μg of T\textsubscript{4} per 100 ml of serum.

Thyroxine-binding by TBPA was evident in all specimens subjected to electrophoresis in the Tris-maleate buffer system. The thyroxine-binding capacity of TBPA was usually near the normal mean of approximately 120 μg of T\textsubscript{4} per 100 ml of serum.

\textsuperscript{9} Values described for thyroxine-binding by TBG and TBPA represent the means of those found during electrophoretic analysis of at least 3 specimens obtained during each treatment period.

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{fig1.png}
\caption{The effect of inguinal herniorrhaphy and of diethylstilbestrol on the binding of thyroxine in the serum of a patient with decreased thyroxine-binding globulin (TBG). Postoperative serum obtained 2 days after surgery. Serum obtained during therapy (obtained after 5 weeks of treatment with diethylstilbestrol, 30 mg daily, p.o.). Curves shown are scans of I\textsuperscript{131}-labeled thyroxine added in the indicated concentrations to sera subjected to electrophoresis in Tris-maleate buffer, pH 8.6. The small peak seen in the TBG zone during estrogenic therapy was not evident in other samples obtained during the same treatment period.}
\end{figure}
(2), but varied with the patient's clinical state, declining to subnormal values during exacerbations of congestive heart failure or in the period following emergency inguinal herniorrhaphy. A zone of radioactivity corresponding in electrophoretic migration to that of serum albumin was the only other area in which added I\(^{131}\)-labeled T\(_4\) was localized.

As previously reported (11, 12), I\(^{131}\)-labeled triac and tetrac were bound primarily to TBPA and secondarily to albumin during electrophoresis in either Tris-maleate or veronal buffers. At standard concentrations of added labeled compounds, the percentile distribution of the derivatives between these two proteins in the patient's serum, obtained during the control state, was not appreciably different from normal.

During the administration of estrogen, prednisolone, or sodium-L-thyroxine, appreciable or consistent differences in thyroxine binding by TBG, as compared with those found during the control state, could not be discerned; the small radioactive peak seen in Figure 2 was not found in other samples obtained during the same treatment period. The thyroxine-binding capacity of TBPA was slightly reduced during administration of diethylstilbestrol, but was unaffected by treatment with prednisolone and L-thyroxine.

Red blood cell uptakes of labeled hormones (Table I). In vitro uptakes of labeled T\(_4\) and T\(_3\) from the patient's plasma by red blood cells were greatly increased in the basal state and were not appreciably altered during the several treatment periods. The red blood cell uptake of labeled triac was normal during both the basal state and the administration of diethylstilbestrol, but underwent a pronounced increase during the immediate postoperative period.

Thyroxine metabolism in vivo (Table I). The peripheral metabolism of I\(^{131}\)-labeled T\(_4\), measured while the patient was in the basal state, was grossly abnormal (9, 13). Fractional rate of turnover of administered hormone was rapid, 23.1 per cent per day. The T\(_4\) distribution space was moderately increased when related to total body weight (0.22 L per kg), and hormonal clearance

---

**FIG. 2.** THE EFFECT OF DIETHYLSTILBESTROL ON THE PROTEIN BINDING AND PERIPHERAL TURNOVER OF THYROXINE IN A PATIENT WITH DECREASED TBG. Thyroxine turnover studies during estrogenic therapy begun on Day 41 of treatment with 30 mg of diethylstilbestrol daily, p.o. Distribution of I\(^{131}\) labeled thyroxine (0.4 \(\mu\)g per 100 ml) in sera obtained during each thyroxine turnover study and subjected to electrophoresis in Tris-maleate buffer, pH 8.6. The small peak of radioactivity seen in the TBG zone during estrogenic therapy was not evident in other samples obtained during the same treatment period.
rate was greatly augmented (3.05 L per day). Because of the subnormal PBI, however, total hormonal disposal, 61.0 μg of iodine per day, was within normal limits. The extrathyroidal organic iodine pool was greatly reduced.

Radioiodine appeared in the urine more rapidly than normal after administration of labeled thyroidine. Butanol extractions of urine on the third, fourth, and fifth daily collections revealed that 3.3, 4.6, and 4.0 per cent, respectively, of urinary 131I was soluble in butanol and relatively insoluble in aqueous alkali; these values were similar to those found in normal individuals (14). Calculated values for the ultimate urinary and fecal excretion of 131I were 74.3 and 25.7 per cent of the administered dose, respectively. Both fecal clearance (0.78 L per day) and degradative clearance of T4 (2.27 L per day) were greatly increased, but the daily fecal excretion rate and daily degradation rate were within normal limits.

Neither the peripheral metabolism of T₄ nor the concentration of hormonal iodine in the plasma was significantly altered by the prolonged administration of large doses of diethylstilbestrol. Neither prednisolone nor thyroid-suppressing doses of sodium-L-thyroxine increased the PBI.

**DISCUSSION**

There is general agreement that TBG serves as a rate-regulating factor in the peripheral metabolism of T₄. TBG, like other proteins with which T₄ is associated, is thought to limit the concentration of free or unbound hormone, thereby retarding its cellular penetration and decreasing the proportion of extrathyroidal hormone that is affixed to the cell. If, then, a constant proportion of the hormone within cellular confines is metabolized per unit time, the fractional rate of disposal of the entire peripheral pool of hormone will vary inversely with the binding activity of TBG (2, 3, 4-6).

### Table I

**Values for several aspects of thyroid hormone economy in normal subjects and in a patient with idiopathic decrease in thyroxine-binding globulin**

<table>
<thead>
<tr>
<th>Function</th>
<th>Normal values†</th>
<th>Control</th>
<th>Diethylstilbestrol</th>
<th>Prednisolone</th>
<th>Postoperative</th>
<th>L-thyroxine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thyroid I³¹ uptake (% dose)</td>
<td>20–50</td>
<td>28</td>
<td>26</td>
<td>6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Basal metabolic rate</td>
<td>−15+15</td>
<td>+3</td>
<td>+6</td>
<td>−1</td>
<td>+4</td>
<td></td>
</tr>
<tr>
<td>Serum cholesterol (mg %)</td>
<td>150–270</td>
<td>210</td>
<td>204</td>
<td>198</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Protein-bound iodine (μg %)</td>
<td>4.0–8.0</td>
<td>2.0</td>
<td>2.1</td>
<td>2.1</td>
<td>1.8</td>
<td>2.3</td>
</tr>
<tr>
<td>Thyroxine space (TDS; L)</td>
<td>9.4 ± 2.0</td>
<td>13.2</td>
<td>15.7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Turnover rate (k; %/day)</td>
<td>10.6 ± 0.9</td>
<td>23.1</td>
<td>19.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Clearance rate (C; L/day)</td>
<td>1.0 ± 0.3</td>
<td>3.05</td>
<td>3.11</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Organic iodine pool (ETT; μg I)</td>
<td>508 ± 146</td>
<td>264</td>
<td>329</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Daily hormone disposal (D; μg I/day)</td>
<td>53.6 ± 16.5</td>
<td>61.0</td>
<td>65.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fecal fraction (Fₚ₀; % dose)</td>
<td>36.9 ± 9.2</td>
<td>25.7</td>
<td>27.2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urinary fraction (Uₚ₀; % dose)</td>
<td>73.1 ± 9.3</td>
<td>74.3</td>
<td>72.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fecal clearance (L/day)</td>
<td>0.27 ± 0.11</td>
<td>0.78</td>
<td>0.85</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Degradative clearance (L/day)</td>
<td>0.73 ± 0.21</td>
<td>2.27</td>
<td>2.26</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fecal excretion (μg I/day)</td>
<td>14.6 ± 6.8</td>
<td>15.6</td>
<td>17.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Degradation rate (μg I/day)</td>
<td>39.0 ± 12.7</td>
<td>45.3</td>
<td>47.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hormonal binding studies (2, 36)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TBG capacity (μg thyroxine/100 ml)</td>
<td>18–25</td>
<td>±‡</td>
<td>±</td>
<td>120</td>
<td>18</td>
<td>121</td>
</tr>
<tr>
<td>TBPA capacity (μg thyroxine/100 ml)</td>
<td>90–160</td>
<td>136</td>
<td>85</td>
<td>150</td>
<td>105</td>
<td>120</td>
</tr>
<tr>
<td>RBC thyroxine uptake (%)</td>
<td>0.6–0.9</td>
<td>1.5</td>
<td>1.5</td>
<td>1.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>RBC triiodothyronine uptake (%)</td>
<td>5.0–8.5</td>
<td>13.2</td>
<td>12.8</td>
<td>14.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>RBC triiodothyroacetic acid uptake (%)</td>
<td>1.2–2.1</td>
<td>1.5</td>
<td>1.9</td>
<td>2.9</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*For a description of functions and abbreviations shown under thyroidine turnover studies, see Appendix. For details regarding the several treatment periods, see text.
† Values presented are normal range or mean ± standard deviation. Normal values are those presented in previous studies or calculated from data contained therein, and were obtained by methods similar to those employed in Patient F.B. Numbers in parentheses provide references to these sources.
‡ ± indicates traces of TBG inconstantly seen, and, when demonstrable, present in concentrations too low to permit calculation of thyroxine-binding capacity.
IDIOPATHIC DECREASE OF THYROXINE-BINDING GLOBULIN

This hypothesis is largely based on inferential evidence, consisting of a series of correlations between changes in extracellular thyroxine-TBG interactions and changes in the metabolism of the hormone, either in vitro or in vivo. Thus, the uptake of T₄ by cellular systems in vitro varies inversely with the concentration of TBG and directly with the concentration of hormone in the suspending medium (15). In vivo correlations have mainly required the use of pharmacological agents to alter the thyroxine-binding capacity of TBG, associated effects on the peripheral metabolism of labeled T₄ being then determined. Diethylstilbestrol and natural estrogens, for example, increase the binding capacity of TBG (13, 16) and decrease the fractional rate of turnover of the hormone (13, 17), while methyltestosterone produces converse effects (18). These in vivo observations, while consistent with the major hypothesis, have not been entirely conclusive because of the possibility that the agents employed might directly affect cellular mechanisms for the disposal of hormone. This seemed especially possible in the case of diethylstilbestrol, in view of reports that this compound inhibits the deiodination of T₄ both by intact cellular preparations and by a partially purified thyroxine-deiodinase (19, 20).

More compelling evidence in favor of the postulated function of TBG has been afforded by the recently described patient in whose plasma there was found an apparently idiopathic increase in TBG (21). This abnormality was accompanied by changes in the peripheral metabolism of T₄ similar to those found in association with the increased TBG induced by estrogens (13). No gross evidence of endogenous excess of estrogens was evident in the male patient described. However, values for urinary estrogens were rather high and those for urinary gonadotropin rather low. These findings, together with the familial nature of the disorder, raised the possibility that some inherited disturbance in the secretion or metabolism of estrogens may have produced the increase in TBG which this patient manifested.

In the present patient, the apparently coincidental association of testicular hypofunction with virtual absence of TBG in the plasma afforded an unusual opportunity to complement existing data concerning the function of TBG. In view of the theoretical objections to the in vivo studies described above, it appeared desirable to obtain conclusive evidence that the abnormality in TBG displayed by this patient was not due to disturbances in known hormonal or metabolic factors which might, in themselves, influence the metabolism of T₄.

This patient’s distinct hypogonadism excluded excessive secretion of normal androgens as the cause of decreased TBG. It appeared possible, however, that the patient’s abnormal testes were secreting an unusual, nonandrogenic steroid capable of decreasing TBG. Apparently the capacity of certain steroids to decrease TBG is not a function of their androgenic potency, since nor-ethandrolone, a steroid with relatively more anabolic than androgenic activity (22), has proved capable of inducing pronounced reductions in TBG within a period as short as 1 week (23). During this time fractional peripheral turnover of T₄ was accelerated, but androgenic effects were not evident, even in hypogonadal patients (23). The large doses of diethylstilbestrol administered to the present patient produced gynecomastia and edema, suppressed gonadotropin production, and should have been adequate to suppress any lingering function that his testes may have retained. Nevertheless, diethylstilbestrol failed to restore the binding capacity of TBG to normal, and, indeed, any increase in TBG that may have been induced by this agent was too small to be recognized consistently during electrophoretic analyses.

Finally, it seems unlikely that an androgen-like steroid of adrenocortical origin was responsible for the decrease in TBG, since prolonged adrenocortical suppression failed to influence the abnormality in hormonal binding. The patient with decreased TBG reported by Tanaka and Starr had a history of hepatitis and displayed abnormal cephalin flocculation and thymol turbidity tests. In the present patient there was no clinical or laboratory evidence of hepatic or renal disease. Therefore, currently recognized causes of decreased TBG appear to have been excluded in the present case.

The abnormalities in the peripheral metabolism of T₄ which accompanied the decrease in TBG in this patient were consonant with the postulated function of TBG. The greatly increased red

---

17a-ethyl-17-hydroxynorandrostenone.
As would therefore be expected, the $T_4$ distribution space was moderately increased in vivo and the fractional turnover of hormone was greatly accelerated. Accordingly, total hormonal clearance rate was markedly increased. However, the PBI was abnormally low, with the result that total hormonal disposal was well within the normal range.

Studies of urinary $I^{131}$ and estimates of fecal excretion made possible an assessment of the general metabolic pathways by which the increased fractional rate of turnover was effected. Although fecal clearance of $T_4$ was increased several-fold, a finding in accord with earlier evidence that protein binding of $T_4$ limits its gastrointestinal excretion (24, 25), this increase accounted for only a small fraction of the increase in total $T_4$ clearance which the patient displayed. In the presence of an increased proportion of unbound hormone in the plasma, an increased renal clearance of $T_4$ would also be expected (14). Normally, however, this route of disposal contributes so little to total hormonal clearance that an enormous increase in this function would have been required to contribute significantly to the abnormality in hormonal turnover (26, 27). Measurements of the butanol-extractable $I^{131}$ in the patient's urine confirmed the relative unimportance of urinary excretion of $T_4$ as a factor in the urinary excretion of $I^{131}$ and in total hormonal disposal. Thus, increased hormonal turnover was due, in greatest part, to increased disposal of $T_4$ by those metabolic pathways which ultimately lead to hormonal deiodination.

It seemed important to exclude the possibility that this patient's subnormal PBI was not due to the abnormality in hormonal binding, but rather reflected the secretion of some thyroid hormone, like $T_3$, which, in amounts sufficient to maintain a normal metabolic state, does not contribute greatly to the PBI (28). If this were the case, then the metabolism of labeled $T_4$ would not have accurately reflected the metabolism of endogenous hormone. However, if $T_4$ were indeed the major hormone in the patient's plasma, then doses of L-thyroxine adequate to suppress endogenous thyroid function should have restored the PBI to normal. Since they did not, it seems likely in the present patient, as is normally the case (29), that $T_4$ constituted the major secretory product of the thyroid gland and the major constituent of the PBI. Calculated values for hormonal disposal were therefore validated.

The present association of normal thyroid function and metabolic status with normal values for daily $T_4$ disposal and deiodination, despite a subnormal PBI, supplements previous evidence, reviewed elsewhere (30), that both the secretion of thyrotropin and the metabolic state of the patient are more closely related to the quantity of hormone available to the tissues than to the total concentration at which they are delivered. The concentration of unbound hormone in the plasma, could it be readily determined, would likely constitute a better index of metabolic state than does the total PBI.

The failure of diethylstilbestrol to alter the kinetics of $T_4$ metabolism in this patient indicates that, in man, in contradistinction to certain cellular preparations in vitro, this agent apparently does not directly affect the cellular degradation of $T_4$. It therefore follows that the changes in $T_4$ metabolism that accompany the administration of estrogen to patients with normal TBG are indeed due to the resulting increases in TBG. Furthermore, the present findings support the belief that the changes in $T_4$ metabolism induced by androgenic hormones are not due to a direct cellular effect of the androgens, but rather to the associated decrease in TBG. Thus, the data lend considerable weight, both directly and indirectly, to the postulated function of TBG, described above. They do not, however, conclusively prove the hypothesis. Alternative interpretations of the function of TBG in the light of the present and previous observations concerning the metabolism of $T_4$ may also be envisioned (2). Furthermore, one cannot logically exclude the possibility that the abnormalities in $T_4$ metabolism which this patient displayed arose, not from the decreased TBG, but from unrelated abnormalities in the cellular metabolism of the hormone.

In the absence of significant hormonal binding by TBG, an unusually large share of the transport of $T_4$ must devolé upon other proteins; notably TBPA (31). However, a consideration of the
possible physiological role of TBPA in the present patient, or indeed, in general, is beyond the scope of this report. While, in a number of circumstances, correlated changes in thyroxine binding by TBPA and in $T_4$ metabolism indicate that this protein contributes significantly to hormonal binding in vivo (2, 12), other evidence suggests that it may not (32, 33). Nevertheless, hormonal binding by TBPA seems adequate to explain several phenomena observed in the present studies. Thus, the normal values for thyroxine binding by TBPA found in the patient’s serum when he was in a basal state correlated well with normal values for the red blood cell uptake of triac, since this derivative is primarily bound to TBPA, and not to TBG (11, 12). Similarly, the increased red blood cell uptake of triac obtained when the patient was in the post-operative period is probably explained by the reduced hormonal binding by TBPA which occurred at that time, a change similar to that found previously in sera from a large number of patients with other nonthyroidal illnesses (34). Subsequent studies in a large number of patients have indicated that the in vitro uptake of deaminated derivatives of $T_4$ and $T_3$ by an adsorptive particulate system varies inversely with, and is a convenient means of estimating, the thyroxine-binding capacity of TBPA (35).

As was the case in the patient reported by Tanaka and Starr (4), the defect in TBG in the present patient was associated with no gross abnormalities in the concentration of the major electrophoretically distinguishable groups of plasma proteins. Of the many transport functions of the plasma proteins only the transport of iron was assessed, and this proved to be normal. Thus, it is uncertain whether the abnormality described here is an isolated defect. In neither patient with this disorder has it been possible to ascertain whether the decrease in TBG was congenital or acquired, or whether it represented an inherited abnormality. The male patient with absent TBG described by Tanaka and Starr had no evidence of cardiac disease (4). Consequently, it is uncertain whether the congenital cardiac lesion in the present patient with decreased TBG represents a related abnormality or merely a fortuitous association.

Although the defect in the present patient has been referred to as a decrease in TBG, the concentration of the protein itself was not measured. Conceivably, decreased hormonal binding could have resulted from the presence of a highly effective inhibitor of thyroxine binding or from a minor structural abnormality in the protein that impaired the attachment of $T_4$. More likely, however, the abnormality described in this paper represents, as in other dysproteinemias (36), a decrease in the concentration of the protein itself. Whether, if this be true, the virtual absence of TBG from this patient’s plasma is due to excessive destruction or defective protein synthesis, as is the case in agammaglobulinemia and analbuminemia (36), can not be determined at present. Resolution of these and other intriguing questions concerning the function and metabolism of TBG await ample supplies of the purified protein.

**SUMMARY**

1. Clinical features have been described of a euthyroid male patient with postoperative primary hypogonadism and a congenital atrial septal defect, whose plasma was virtually devoid of thyroxine binding by the thyroxine-binding globulin (TBG).

2. Prolonged administration of large doses of synthetic estrogen or adrenal corticosteroid failed to alter significantly the binding activity of TBG, suggesting that the abnormality in this protein was not produced by an androgen of testicular or adrenocortical origin.

3. This seemingly idiopathic decrease in TBG was associated with pronounced abnormalities in the peripheral metabolism of thyroxine. Both the volume of distribution and fractional rate of turnover of $I^{131}$-labeled thyroxine were increased; hormonal clearance rate was markedly augmented. However, the serum protein-bound iodine was subnormal, with the result that daily disposal of hormone by both excretory and degradative routes was within normal limits.

4. In contrast to its action in normal individuals, in whom it induces an increased binding activity of TBG, estrogenic therapy had no significant effect upon the concentration or peripheral turnover of thyroxine in this patient.

5. The several findings in this patient are viewed as consistent with the hypothesis that TBG regulates the peripheral metabolism of thyroxine by
limiting its cellular penetration and hence its peripheral metabolism and action.

APPENDIX

In this manuscript and Table I the following terms have been employed. These were described in detail in an earlier communication (9).

TDS, thyroxine distribution space; the virtual volume of body fluids through which exchangeable thyroxine would be distributed were it present throughout at the same concentration at which it exists in the plasma.

PBI, concentration of hormonal iodine in the plasma, and, by definition, the geometrical mean concentration of hormonal iodine in the TDS; assumed to represent solely iodine in thyroxine.

ETT, extrathyroidal thyroxine or the organic iodine pool; the total quantity of thyroxine within the TDS, in terms of its content of iodine.

$k$, fractional rate of turnover of thyroxine; the proportion of ETT removed from the TDS per unit time.

$T_1$, thyroxine half-time; the time required for one-half of the ETT to be removed from the TDS.

$C$, thyroxine clearance rate; the volume of plasma containing an amount of thyroxine equal to that removed from the TDS per unit time. Defined as “volume turnover, $V$” in an earlier communication (9).

$D$, hormonal disposal rate; the quantity of hormone removed from the TDS per unit time, in terms of its content of iodine.

$F_{max}$, the proportion of $D$ accounted for by fecal excretion of hormone; $U_{max}$, the proportion of $D$ accounted for by urinary excretion of organic iodine and iodide released during hormonal deiodination; fecal clearance, the volume of plasma that contains a quantity of hormone equal to that lost in the feces per unit time; degradative clearance, the volume of plasma containing a quantity of hormone equal to that removed by nonexcretory pathways per unit time. As calculated, degradative clearance includes the renal clearance of thyroxine, but the latter comprises a negligible proportion of the total; fecal excretion, the quantity of hormone excreted in the feces per unit time; degradation rate, the quantity of hormone removed by nonexcretory pathways per unit time.

$$ETT = PBI \times TDS, \quad C = TDS \times k, \quad D = C \times PBI = ETT \times k, \quad T_1 = 0.693/k, \quad \text{Fecal clearance} = C \times F_{max}, \quad \text{Degradative clearance} = C \times U_{max}, \quad \text{Fecal excretion} = \text{fetal clearance} \times PBI = D \times F_{max}, \quad \text{Degradation rate} = \text{degradative clearance} \times PBI = D$ - \text{fetal excretion}.$$

ACKNOWLEDGMENTS

The author is indebted to Miss Patricia Eveleth and Miss Rebecca Feretos for valuable technical assistance.

REFERENCES


18. Federman, D. D., Robbins, J., and Rall, J. E. Effects of methyl testosterone on thyroid function, thy-