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Serum Thyrotropin and Triiodothyronine Levels in Levothyroxine-treated Patients

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Abstract

Context: Small adjustments in levothyroxine (LT4) dose do not appear to provide clinical benefit despite changes in thyrotropin (TSH) levels within the reference range. We hypothesize that the accompanying changes in serum total triiodothyronine (T3) levels do not reflect the magnitude of the changes in serum TSH.

Objective: This work aims to characterize the relationships of serum free thyroxine (FT4) vs T3, FT4 vs TSH, and FT4 vs the T3/FT4 ratio.

Methods: This cross-sectional, observational study comprised 9850 participants aged 18 years and older treated with LT4 from a large clinical database from January 1, 2009, to December 31, 2019. Patients had been treated with LT4, subdivided by serum FT4 level. Main outcome measures included model fitting of the relationships between serum FT4 vs TSH, FT4 vs T3, and FT4 vs T3/FT4. Mean and median values of TSH, T3, and T3/FT4 were calculated.

Results: The relationships T3 vs FT4 and TSH vs FT4 were both complex and best represented by distinct, segmented regression models. Increasing FT4 levels were linearly associated with T3 levels until an inflection point at an FT4 level of 0.7 ng/dL, after which a flattening of the slope was observed following a convex quadratic curve. In contrast, increasing FT4 levels were associated with steep declines in TSH following 2 negative sigmoid curves. The FT4 vs T3/FT4 relationship was fit to an asymptotic regression curve supporting less T4 to T3 activation at higher FT4 levels.

Conclusion: In LT4-treated patients, the relationships between serum FT4 vs TSH and FT4 vs T3 across a range of FT4 levels are disproportionate. As a result, dose changes in LT4 that robustly modify serum FT4 and TSH values may only minimally affect serum T3 levels and result in no significant clinical benefit.

Key Words: hypothyroidism, thyrotropin, levothyroxine, triiodothyronine

Abbreviations: D2, type II deiodinase; FT4, free thyroxine; IQR, interquartile range; LT4, levothyroxine; TPO, thyroid peroxidase; T3, total triiodothyronine; TSH, thyrotropin.

The rationale for the treatment of hypothyroidism with levothyroxine (LT4) monotherapy is that thyroxine (T4) is rapidly absorbed into the circulation and undergoes outer ring deiodination to the biologically active hormone triiodothyronine (T3). This was elegantly demonstrated in the early 1970s (1, 2) and taken as an indication that LT4 alone could be used to treat patients with hypothyroidism. In thyroidectomized patients or those with no functional thyroid gland, the deiodinase pathways constitute the only source of T3. Two distinct deiodinase pathways exist for the conversion of T4 to T3: type I deiodinase (D1) and type II deiodinase (D2). Studies in which the D1 pathway was inhibited with propylthiouracil revealed that D2 is responsible for approximately 80% of the circulating T3 levels in LT4-treated thyroidectomized patients (3). Type III deiodinase (D3) affects circulating T3 levels by inactivating T3 to diiodothyronine (T2), and normally converts only a small fraction of T4 into the inactive hormone reverse T3 (4).

According to clinical guidelines for the treatment of hypothyroidism, the dose of LT4 is adjusted for each patient based on serum thyrotropin (TSH) levels (5, 6). The ideal dose of LT4 is achieved when serum TSH levels have been restored to the normal reference range. This occurs via a feedback mechanism triggered by both circulating T3 and by T3 locally converted from T4 by D2 in the pituitary gland and the medial basal hypothalamus (7).

For decades it has been assumed that the normalization of serum TSH in LT4-treated patients occurs simultaneously with the normalization of serum T3 levels and the resolution of symptoms. Accordingly, several clinical guidelines do not recommend routine measurement of serum T3 in the follow-up of patients with hypothyroidism (8). However, observational

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studies involving thousands of patients taking LT4 show that normal serum TSH levels coexist with relatively lower serum T3 levels and T3/T4 ratio, including about 15% of the patients who have serum T3 levels below the reference range (9-19). Fewer studies failed to detect a difference in serum T3 levels but confirmed an elevated serum T4/T3 ratio in LT4-treated patients (20-24). Likely due to these observations, it is commonplace among physicians to recommend higher doses of LT4 in response the patients with hypothyroid symptoms and TSH levels in the upper half of the normal range (25, 26). These practice patterns persist despite a lack of evidence that patients benefit from LT4 dose adjustments within the normal range (27-29). There is now renewed attention on serum T3 levels during therapy with LT4, specifically focusing on the relationship between serum T4 and T3 levels in these patients and the relationship between changes in the LT4 dose (and serum T4 levels) and changes in serum T3 levels (30).

We believe that the relationship between free T4 (FT4) and T3 levels is characterized by small, incremental changes in T3 in response to increasing FT4 levels in LT4-treated patients. This is in contrast to TSH levels, which decrease sharply in response to increases in FT4. In rodents, this is explained by variable turnover rates of D2, which normally accelerates during the T4 activation process to T3 (31). This occurs through D2 ubiquitination and the subsequent destruction in the proteasome system (32, 33). However, this process varies throughout the body. It occurs more slowly in the hypothalamus and the pituitary gland compared to peripheral tissues. This heterogeneity sets the stage for a possible disconnect between serum TSH and T3 levels in LT4-treated patients and has implications for TSH regulation and the treatment of patients with hypothyroidism (34).

The purpose of this cross-sectional analysis is to study the relationships between (1) the FT4 vs TSH serum levels; and (2) the serum FT4 vs total T3 levels in a large data set of patients treated with LT4 via growth curve modeling. We propose that differences in central (hypothalamus-pituitary unit) and peripheral (all other tissues) D2 activity are the primary drivers of a disconnect between the response of T3 and TSH to changes in FT4 in LT4-treated patients.

Material and Methods

This is a cross-sectional observational study including a large, Brazil-based, commercial database from January 1, 2009, to December 31, 2019 (Fleury Group, São Paulo, Brazil) (35). Deidentified data were pulled under institutional review board approval number NP-458. Additionally, this secondary analysis was exempted from institutional review board review at the University of Chicago.

Data Source

The Fleury Group database, containing the results of a 10-minute unique patient interview conducted by a medical assistant followed by a clinical laboratory encounter in 6 Brazilian states, captured approximately 100 million people. Clinical data are entered manually after an attendant creates a new entry and asks the patient about medications in use. The information is typed in a free text format and stored in a Microsoft SQL database. Information is kept secure through a separate network firewall, accessed only by authorized individuals within the Fleury Group's domains. Data stored in this database have been used previously in several clinical studies (36-39). Clinical data (TSH, FT4, T3) and minimal demographic data (sex, age) from those taking LT4 were deidentified and extracted from the primary data set.

Study Population

All individuals aged at least 18 years for whom thyroid function tests were available were included in the study. Each entry corresponded to a unique patient; only the most recent laboratory data from each patient were retrieved. The commercial names of medications were obtained as previously described (35). Briefly, this was done using local alignment dynamic programming as implemented in the "pairwise-alignment" function from the Biostrings R package (Bioconductor project) (40) using a gap opening penalty of 2 and a gap extension penalty of 0.5. The lists of medications were aligned against each patient's medication string to obtain overlapping data. Individuals whose medication list included LT4 (generic or brand name) were included in the study population. To avoid the potential effects of drug-drug interactions on thyroid function, individuals taking any other medications (including over-the-counter medications) were excluded. Individuals who were pregnant within 12 months of the time of laboratory collection were excluded. Those patients who were significantly overtreated (FT4 levels > 2.5 ng/dL) and those with inappropriately low or normal TSH levels (TSH < 4.5 mIU/L and FT4 < 0.7 ng/dL) were excluded.

Thyroid function assays

Total serum T3 levels were measured using an electrochemoluminescent competitive immunoassay (Roche; reference range 70-200 ng/dL), FT4 serum levels were measured using a chemoluminescent competitive immunoassay (Beckman Coulter; reference range 0.7-1.3 ng/dL), and serum TSH levels were measured using an electrochemoluminometric assay (Roche; reference range 0.45-4.5 mIU/L). Thyroid peroxidase (TPO) antibodies were measured using an electrochemoluminometric assay, radioimmunoassay, or immunofluorimetric (Roche) assay throughout the study period. Total serum T3 levels were used as the measure of T3 because of the lack of accuracy in measuring free T3 using standard immunoassay techniques (41). All laboratory samples were collected in the fasted state, before LT4 administration.

Exposure and outcomes and covariates

Exposure was the utilization of LT4. The FT4 level was considered as an independent variable corresponding to the exposure to LT4. The primary outcomes were serum TSH, T3, and T3/FT4 ratio. Age and sex were covariates in the descriptive analyses.

Statistical methods

Thyroid function tests (TSH, FT4, T3) from all participants were summarized via mean and median due to the nonnormal distributions of thyroid function levels throughout the study population. Five subgroups were formed based on FT4 level: 0.2 to 0.7, 0.8 to 1.0, 1.1 to 1.3, 1.4 to 1.7, and 1.8 to 2.5 ng/dL. Mean values between the subgroups were compared via analysis of variance, and median values were compared via the Kruskal-Wallis test. Descriptive analyses were also conducted per subpopulation stratified by age group (18-50 and > 50 years) and sex.

Due to complex and/or nonlinear relationships between TSH and T3 and FT4, we carefully examined scatterplots and box plots of each outcome by FT4 level before modeling. Due to the skewness of our outcomes, we plotted the median and upper and lower quartiles at each FT4 value.

To capture the curvature of log TSH vs FT4, as done previously (42), and also confirmed by our plot, we used segmented nonlinear quantile regression. The breakpoint between the 2 segments was at 0.7 ng/dL, the low end of the normal reference range of FT4. In each segment, log TSH was fit to a sigmoid curve (with the scaled Weibull cumulative distribution function) in 4 parameters (A-D):

$$\ln(TSH) = A + B \times e^{(-e^{(C)} \times \boldsymbol{FT4}^{D})}.$$

Similarly, to model T3, the model had 2 segments at the same break point as modeling log TSH. However, the distribution of T3 values was found to be approximately normal, thus the quantile regression approach was not used. In the first segment (FT4 0.2-0.7 ng/dL), T3 was fit in a linear regression with 2 parameters (E and F):

$$T3 = E + F \times \boldsymbol{FT4}.$$

In the second segment (FT4 0.7-2.5 ng/dL), T3 was fit to a quadratic regression with 3 parameters (G-I):

$$T3 = G - H \times \boldsymbol{FT4} + I \times \boldsymbol{FT4}^2.$$

To model the T3/FT4 ratio, we used the asymptotic regression model with 3 parameters (J-L) across all FT4 values:

$$\frac{T3}{FT4} = J + (K + J) \times e^{-e^{L} \times \boldsymbol{FT4}}.$$

All standard linear and self-starting nonlinear model types available in the built-in R "stats" package (43) were considered during model fitting (eg, asymptotic regression, logistic model, Gompertz growth curve, Weibull growth curve) through a progression from fewest to most parameters up to 4 parameters. All models were fit based on the median value at each distinct FT4 level to mediate the effect of potential outliers. Final model types were selected based on best visual fit, goodness-of-fit statistics (eg, R^2 [linear], pseudo R^2 [nonlinear], Akaike information criterion), and using the least number of parameters for best fit. Due to only small differences being observed in the stratified analyses, sex and age were not included as covariates in the thyroid function models. All analyses were conducted in RStudio (version 4.1.2). The R "quantreg" package was used for nonlinear regression in the log TSH model.

Results

Approximately 320 000 (6.0%) patients reported being on LT4 within the Fleury Group database. Of those patients, 69 706 (21.8%) patients had available results for serum TSH, FT4, and T3 (35). Of these, 57 439 patients were excluded because they were taking other medications in addition to LT4. A total of 980 patients were excluded because of pregnancy within 1 year of the index encounter. An additional 1419 were excluded because of missing data, 10 patients were excluded with an FT4 level greater than 2.5 ng/dL, and 8 patients were excluded with an FT4 level less than 0.7 ng/dL and an inappropriately low or normal TSH level (< 4.5 mIU/L). Ultimately, 9850 patients on LT4 were analyzed (Fig. 1). Of those patients included in the final study group, the presence of TPO antibodies was assessed in 4206 (42.7%) patients. Of those patients, 1833 (43.6%) had detectable TPO antibodies (median 296 IU/mL, interquartile range [IQR, 99-965 IU/mL]).

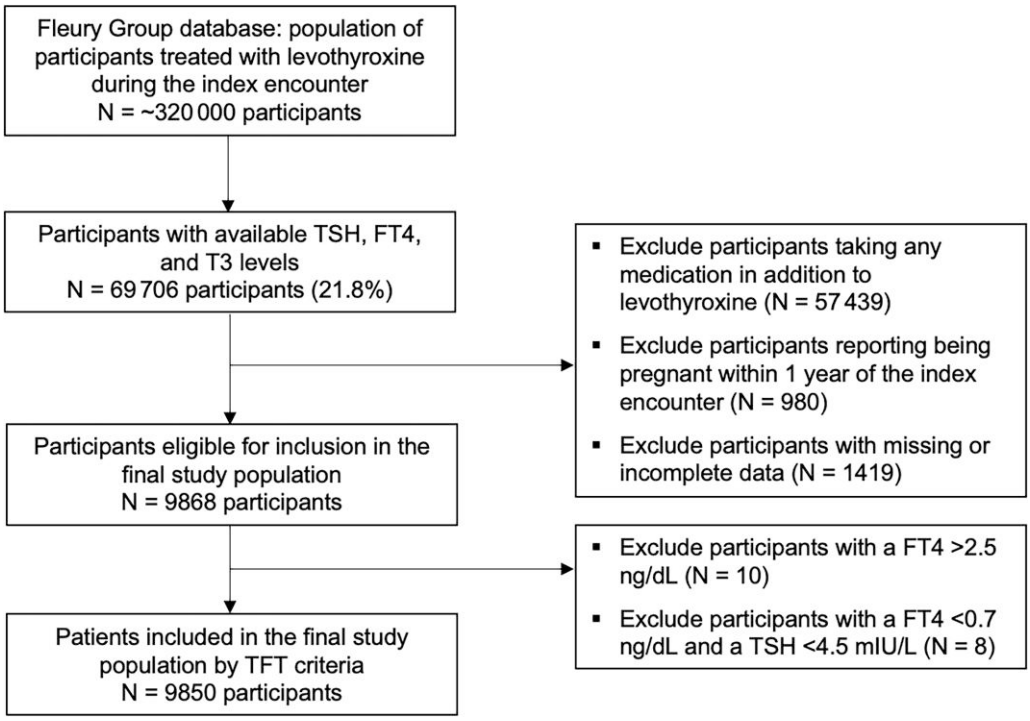

Figure 1. Flowchart of study population selection of participants treated with levothyroxine with available thyroid function tests. FT4, free thyroxine; T3, total triiodothyronine; TFT, thyroid function test; TSH, thyrotropin.

Table 1. Average thyrotropin, total triiodothyronine (T3), and T3/free thyroxine (FT4) values by FT4 range in all participants

All participants	FT4 range, ng/dL					*P*
	0.2-0.7 (n = 745)	0.8-1.0 (n = 5039)	1.1-1.3 (n = 3011)	1.4-1.7 (n = 879)	1.8-2.5 (n = 176)	
TSH, mIU/L						
Mean (SD)	10.8 (23.4)	3.1 (4.1)	1.6 (2.3)	0.54 (1.2)	0.17 (0.6)	< .001
Median (IQR)	3.6 (1.8-7.6)	2.3 (1.2-3.7)	0.96 (0.23-2.2)	0.09 (0.05-0.44)	0.05 (0.05-0.05)	< .001
T3, ng/dL						
Mean (SD)	103.6 (31.5)	102.0 (21.3)	104.5 (20.9)	114.1 (23.5)	127.5 (32.0)	< .001
Median (IQR)	98 (85-116)	99 (88-113)	102 (91-116)	111 (99-127)	126 (109-137)	< .001
T3/FT4						
Mean (SD)	158.4 (48.8)	112.9 (25.7)	89.3 (18.2)	76.5 (15.7)	64.4 (15.0)	< .001
Median (IQR)	148.3 (128.3-175.7)	109.0 (96.0-126.3)	87.5 (76.7-99.1)	75.6 (66.0-85.7)	62.8 (54.0-72.1)	< .001

Median and mean values of TSH, T3, and T3/FT4 in all participants, stratified by FT4 level. Mean values tested via analysis of variance. Median values compared via the Kruskal-Wallis test.
Abbreviations: FT4, free thyroxine; IQR, interquartile range; T3, total triiodothyronine; TSH, thyrotropin.

Overall Study Population Characteristics

The median age of the study population was 42 years (IQR, 37-49 years). A total of 2147 study participants (21.8%) were male. The median FT4, TSH, T3, and T3/FT4 levels were 1.0 ng/dL, 102.0 ng/dL, 1.7 mIU/L, and 105.0, respectively. Median and mean TSH, T3, and T3/FT4 values stratified by FT4 subgroup are summarized in Table 1.

Relationship Between Free Thyroxine vs Thyrotropin

The relationship between FT4 vs TSH (log scale) is depicted in Fig. 2A. Median TSH values for each FT4 value are plotted with upper and lower quartiles. Similar to the previous description in non-LT4 users (42), the model is composed of 2 negative sigmoid curves, with the first corresponding to FT4 values at or below and lower limit of normal (FT4 $\leq$ 0.7 ng/dL), with the second corresponding to all other FT4 values. The formula of each segment estimated via quantile regression is included in Table 2. The model suggests that as FT4 values increase in LT4 users, TSH levels rapidly decline as FT4 enters and exits the normal physiological range, with a relatively stable period within the normal range.

Relationship Between Free Thyroxine vs Total Triiodothyronine

The relationship between FT4 vs T3 is depicted in Fig. 2B. Median T3 values for each FT4 value are plotted with upper and lower quartiles. Similar to the FT4 vs TSH model, this model is separated into 2 segments, with FT4 values above and below 0.7 ng/dL. The formula for each segment is included in Table 2. The first segment is modeled linearly as T3 levels increase steadily until the lower bound of the normal range of FT4 is crossed. Segment 2 is modeled via a convex quadratic curve that illustrates the stability of T3 levels throughout a range of FT4 values that extended to FT4 values near 2.0 ng/dL.

Relationship Between Free Thyroxine (FT4) vs the Total Triiodothyronine/FT4 Ratio

The T3/FT4 ratio was examined given the prior observations that patients treated with LT4 may have a relatively lower level of T3 compared to T4. Shown in Fig. 2C are the plotted median T3/FT4 values at each FT4 value, with upper and lower quartiles. The median T3/FT4 values were fit to an asymptotic regression curve (see Table 2). Unlike the prior models, this model was not segmented, meaning a consistent relationship was observed across all FT4 values. To summarize, the T3/FT4 ratio declined as FT4 increased; however, the rate of decline slowed over the entire FT4 range.

Age and Sex Stratification

As expected, average TSH values decreased significantly across groups stratified from lower to higher FT4 levels (Tables 3 and 4). Across all groups, average T3 levels largely remained near 100 ng/dL except when FT4 values exceeded 1.3 ng/dL. Of note, the median TSH level in the lowest FT4 subgroup was 3.2 mIU/L in women and 5.1 mIU/L in men. In general, average TSH values in men trended higher in each FT4 subgroup compared to women. No major differences were noted in T3 or T3/FT4 values. No major differences in average thyroid function values were noted between those older and younger than 50 years.

Discussion

In this study, we examined the relationships between serum FT4 and TSH, T3, and T3/FT4 levels in LT4-treated patients. As expected, the relationship between serum TSH vs FT4 values was best described by 2 negative sigmoid curves, similar to those of prior studies that demonstrated a complex, nonlinear relationship between log TSH vs FT4 (42, 44, 45). In contrast to the log TSH vs FT4 relationship, the FT4 vs T3 relationship was best defined as linear at low FT4 levels and quadratic at normal and high FT4 levels, highlighted by relatively stable serum T3 levels throughout the FT4 reference range and beyond. This was supported by the FT4 vs T3/FT4 relationship, which

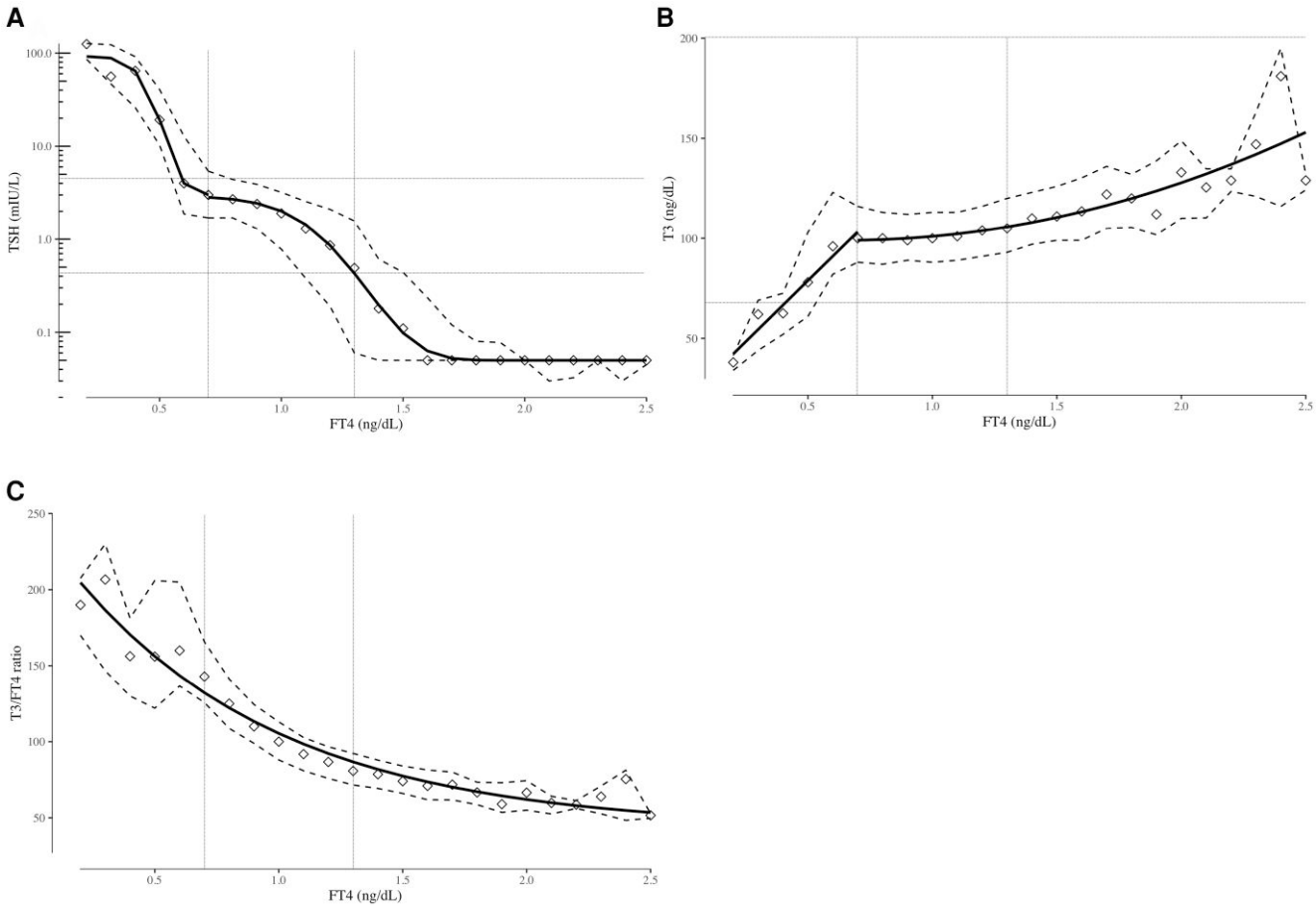

Figure 2. A, Relationship between log TSH and FT4 in levothyroxine-treated individuals. The median TSH value for each FT4 value is represented (◊). The solid curve is derived from 2 negative sigmoid (Weilbull) curves. The 2 curved segments are split at the low end of the normal FT4 reference range (0.7 ng/dL). TSH is represented on a logarithmic scale and FT4 on a linear scale. B, Relationship between T3 and FT4 in levothyroxine-treated subjects. The median T3 value for each FT4 values is represented (◊). The solid curve is derived from a linear segment and a convex quadratic segment. The 2 curved segments are split at the low end of the normal FT4 reference range (0.7 ng/dL). C, Relationship between T3/FT4 and FT4 in levothyroxine-treated individuals. The median T3/FT4 value for each FT4 value is represented (◊). In this case, the solid line is derived from a single asymptotic regression curve. All curve formulas are summarized in Table 2. The upper and lower quartiles in each figure are depicted by the dashed lines. Normal ranges of TSH, FT4, and T3 are indicated by dotted lines (TSH: 0.45-4.5 mIU/L; FT4: 0.7-1.3 ng/dL; T3: 70-200 ng/dL). FT4, free thyroxine, T3, total triiodothyronine; TSH, thyrotropin.

was fit to an asymptotic regression curve, demonstrating that the rate of decline of the T3/FT4 ratio slows as FT4 increases. Similar to those prior studies, we did identify minor differences in thyroid function tests by age and sex.

By examining in parallel the behavior of serum T3 and TSH levels in response to increasing FT4 levels, model data suggest the sharp decline in TSH levels is paired with a steady rise in T3 levels only when FT4 levels are below the normal range (< 0.7 ng/dL). As TSH levels continue to decline rapidly at the upper end of the normal range of FT4 (1.2-1.4 ng/dL), T3 levels remain relatively flat. Thus, an increase in FT4 levels is greatly effective in reducing TSH but not nearly as effective in elevating serum T3 levels. The former depends primarily on the D2-mediated conversion of T4 to T3 in the hypothalamus and pituitary gland, whereas the latter depends on D2-mediated T4 activation throughout the body. Although mediated by the same enzyme, the disconnect between these 2 processes may be attributed to differences in D2 regulation, as described in LT4-treated, thyroidectomized rats (33). In the animal model, D2 activity in the hypothalamus-pituitary unit was only minimally affected by T4, whereas D2 activity (T4 to T3 conversion) was significantly reduced in tissues that contribute to circulating T3. The potential existence of a similar mechanism in humans could explain the comparability of T3 levels over a range of FT4 levels in patients taking LT4.

These findings address an important knowledge gap among clinicians managing patients LT4-treated patients. Across multiple surveys, more than 60% of physicians would recommend increasing LT4 in response to patients with hypothyroid symptoms and a TSH level of 3.9 mIU/L, regardless of whether T3 levels are low or normal (25, 26). We believe the findings of this study, along with those of Walsh et al (27) and Samuels et al (28, 29), refute the idea that increasing doses of LT4 in situations like the aforementioned clinical setting will result in clinical benefit for the patient. The TSH level may suppress, but the evidence would suggest little change in circulating T3 level, which then results in little to no appreciable change in symptoms. An important caveat to this conclusion is that circulating T3 levels remain an indirect marker of thyroid hormone action on tissues, and the optimal T3 level for end-organ function (and ultimately symptoms) is not known.

Table 2. Thyroid function relationships with regression models

Relationship	Segment 1 (FT4 0.2-0.7 ng/dL)		AIC	(pseudo)[a] R^2	Segment 2 (FT4 0.7-2.5 ng/dL)		AIC	(pseudo)[a] R^2
	Regression	Formula			Regression	Formula		
TSH vs FT4	Weibull	$\ln(TSH) = 0.47 + 1.49e^{-e^{4.82} \times FT4^{7.66}}$	−6.44	0.97	Weibull	$\ln(TSH) = -1.30 + 1.76e^{-e^{2.35} \times FT4^{7.23}}$	−77.23	0.99
T3 vs FT4	Linear[b]	$T3 = 17.8 + 122.1 \times FT4$	41.39	0.94	Quadratic	$T3 = 105.5 - 20.29 \times FT4 + 15.71 \times FT4^2$	150.84	0.69
$\frac{T3}{FT4}$ vs FT4[b]	Asymptotic	$\frac{T3}{FT4} = 43.01 + (248.09 + 43.01)e^{-e^{0.17} \times FT4}$	242.04	0.96	–	–		

Formulas of each segment of the regression curves of the relationships between TSH, T3, and T3/FT4 vs FT4. Models were fit to the median values of TSH, T3, and T3/FT4 for each FT4 value. Models were chosen based on visual inspection and goodness of fit parameters. R^2 for the linear segment and pseudo R^2 for the nonlinear segments and the AIC are provided for each segment.

Abbreviations: AIC, Akaike information criterion; FT4, free thyroxine; IQR, interquartile range; T3, total triiodothyronine; TSH, thyrotropin.

[a]Pseudo R^2 was calculated for Weibull and asymptotic regressions.

[b]The T3/FT4 vs FT4 model was fitted over the entire FT4 range (0.2-2.5 ng/dL).

We discourage the conclusion from these findings that suppressed TSH levels below the normal reference range are safe because T3 levels are likely to remain in the normal range. TSH levels, along with signs and symptoms of thyroid hormone excess, remain important markers of overtreatment in hypothyroidism (46). TSH levels outside the normal reference range have been associated with adverse clinical outcomes, including cardiovascular events, fractures, and mortality (47-50). Further research is needed to understand the role of T4 and T3 levels in predicting clinical outcomes in LT4-treated patients, but T4 may play a more direct role in cardiovascular and bone health outcomes despite circulating T3 levels being in the normal range.

The present study is not without limitations. Given that the data were extracted from a large national database, the specific hypothyroidism diagnosis for each participant was not available, and the residual degree of thyroid function within the population is not known. In addition, doses of LT4 were not obtained. The study population excluded patients on medications other than LT4, suggesting that our population was likely younger and healthier than the population of LT4 users overall. These exclusions were used to minimize the potential of drug-drug interaction on thyroid function tests. These results should be interpreted in this context. As a cross-sectional study, thyroid function testing was obtained at a single time point for each participant. Thus, no longitudinal assessments could be made. Thyroid function tests, in particular TSH, are sensitive to acute illness, but as these data were collected as part of a national survey, we believe these effects to be minimal. In this study, the only tissue assessment of thyroid hormone action is of the hypothalamus-pituitary unit (ie, TSH production). The actions of T4 and T3 on other tissues were not evaluated. Finally, it has been observed that the measurement of T3 by standard immunoassay techniques may overestimate T3 levels, in particular at lower concentrations (41, 51). This may affect the characterization of the relationships between FT4 and T3 at the lower FT4 range. However, by using median values, a large sample size, and without interference from other medications, we anticipate the overall effect of this effect to be small. Also, these assays are the most widely available in clinical practice today. Further studies characterizing the relationship between FT4 and T3 (total and free) in LT4-treated patients using more advanced laboratory techniques (including liquid chromatography–tandem mass spectrometry [51]) are warranted.

Conclusion

In LT4-treated patients, the relationships between FT4 vs TSH and FT4 vs T3 are complex, nonlinear, and distinct from each other. In models using cross-sectional data, differences in serum T3 levels were minimal throughout and above the normal FT4 range, whereas TSH levels rapidly decreased both at the lower and upper ends of the normal FT4 range. The coexistence of these patterns supports the hypothesis that thyroid hormone metabolism via D2 is different between the hypothalamus-pituitary unit and peripheral tissues, as previously seen in animal models. These findings argue against the commonly held belief in clinical decision-making that increasing the dose of LT4 can benefit patients with persistent hypothyroid symptoms despite TSH being in the normal range.

Table 3. Average thyrotropin, total triiodothyronine (T3), and T3/free thyroxine (FT4) values by FT4 range in all participants, stratified by age

Age < 50 y	FT4 range, ng/dL					*P*
	0.2-0.7 (n = 589)	0.8-1.0 (n = 3875)	1.1-1.3 (n = 2265)	1.4-1.7 (n = 667)	1.8-2.5 (n = 130)	
TSH, mIU/L						
Mean (SD)	10.3 (21.3)	3.1 (4.3)	1.6 (2.4)	0.55 (1.2)	0.18 (0.6)	< .001
Median (IQR)	3.4 (1.8-7.5)	2.3 (1.1-3.7)	1.0 (0.23-2.2)	0.10 (0.05-0.44)	0.05 (0.05-0.05)	< .001
T3, ng/dL						
Mean (SD)	104.8 (33.0)	102.0 (21.6)	103.9 (20.7)	114.3 (24.0)	128.1 (32.6)	< .001
Median (IQR)	99 (84-117)	99 (88-113)	102 (90-115)	112 (99-127)	126 (111-138)	< .001
T3/FT4						
Mean (SD)	159.6 (50.2)	113.1 (26.2)	88.8 (17.9)	76.3 (16.0)	64.3 (14.9)	< .001
Median (IQR)	148.6 (127.1-177.1)	109.0 (95.6-126.3)	87.3 (76.7-98.2)	75.3 (65.8-85.3)	63.8 (53.9-73.2)	< .001
Age ≥ 50 y	**(n = 156)**	**(n = 1164)**	**(n = 746)**	**(n = 212)**	**(n = 46)**	
TSH, mIU/L						
Mean (SD)	13.0 (30.0)	3.2 (3.6)	1.5 (2.1)	0.49 (1.1)	0.15 (0.6)	< .001
Median (IQR)	4.2 (2.0-9.7)	2.4 (1.3-4.0)	0.9 (0.2-2.2)	0.08 (0.05, 0.4)	0.05 (0.05-0.05)	< .001
T3, ng/dL						
Mean (SD)	99.2 (24.8)	102.0 (20.4)	106.4 (21.5)	113.5 (22.0)	125.8 (30.4)	< .001
Median (IQR)	97.0 (85.8-113.0)	101.0 (89-112)	105 (91-119)	110.0 (97.8-127.0)	124.5 (104.3-136.5)	< .001
T3/FT4						
Mean (SD)	153.9 (43.2)	112.3 (23.7)	90.8 (19.0)	76.9 (15.0)	64.7 (15.4)	< .001
Median (IQR)	144.3 (128.6-170.4)	110.0 (96.7-125.0)	89.2 (77.7-101.4)	75.7 (66.5-86.5)	59.5 (54.8-70.8)	< .001

Median and mean values of TSH, T3, and T3/FT4 in all participants, stratified by FT4 level and age younger than or older than 50 years. Mean values tested via analysis of variance. Median values compared via Kruskal-Wallis test. Reference ranges: TSH: 0.45 to 4.5 mIU/L; FT4: 0.7 to 1.3 ng/dL; T3: 70 to 200 ng/dL.
Abbreviations: FT4, free thyroxine; IQR, interquartile range; T3, total triiodothyronine; TSH, thyrotropin.

Table 4. Average thyrotropin, total triiodothyronine (T3), and T3/free thyroxine (FT4) values by FT4 range in all participants, stratified by sex

Women	FT4 range, ng/dL					*P*
	0.2-0.7 (n = 626)	0.8-1.0 (n = 3954)	1.1-1.3 (n = 2295)	1.4-1.7 (n = 693)	1.8-2.5 (n = 135)	
TSH, mIU/L						
Mean (SD)	9.2 (19.2)	2.8 (3.5)	1.4 (2.0)	0.45 (0.9)	0.15 (0.5)	< .001
Median (IQR)	3.2 (1.7-6.7)	2.1 (1.1-3.4)	0.8 (0.18-2.0)	0.08 (0.05-0.39)	0.05 (0.05-0.05)	< .001
T3, ng/dL						
Mean (SD)	104.0 (32.2)	102.4 (22.2)	104.5 (21.2)	114.0 (24.1)	127.8 (34.2)	< .001
Median (IQR)	98 (84-117)	100 (88-113)	102 (91-116)	110 (98-127)	124 (108-137.5)	< .001
T3/FT4						
Mean (SD)	158.5 (48.7)	113.5 (26.7)	89.2 (18.4)	76.3 (16.1)	64.4 (15.4)	< .001
Median (IQR)	148.3 (127.1-177.0)	109.0 (95.6-127.5)	87.5 (76.7-99.1)	75.0 (65.3-85.0)	62.7 (53.7-73.5)	< .001
Men	**(n = 119)**	**(n = 1085)**	**(n = 716)**	**(n = 186)**	**(n = 41)**	
TSH, mIU/L						
Mean (SD)	19.3 (37.6)	4.2 (5.6)	2.2 (3.0)	0.85 (1.8)	0.26 (0.9)	< .001
Median (IQR)	5.1 (3.2-14.3)	3.1 (1.8-5.0)	1.5 (0.46-3.1)	0.22 (0.05-0.8)	0.05 (0.05-0.08)	< .001
T3, ng/dL						
Mean (SD)	101.6 (27.9)	100.8 (17.6)	104.5 (20.1)	114.5 (21.2)	126.5 (23.7)	< .001
Median (IQR)	100.0 (87-114)	99.0 (89-111)	103 (91-116)	115.0 (102.0-127.8)	127 (113-135)	< .001
T3/FT4						
Mean (SD)	157.9 (49.7)	110.9 (21.2)	89.5 (17.7)	76.9 (14.4)	64.4 (13.6)	< .001
Median (IQR)	150.0 (132.3-170.0)	109.0 (96.0-123.3)	88.3 (77.5-99.1)	76.9 (67.1-87.3)	63.8 (57.3-70.6)	< .001

Median and mean values of TSH, T3, and T3/FT4 in all participants, stratified by FT4 level and sex. Mean values tested via analysis of variance. Median values compared via Kruskal-Wallis test. Reference ranges: TSH: 0.45 to 4.5 mIU/L; FT4: 0.7 to 1.3 ng/dL; T3: 70 to 200 ng/dL.
Abbreviations: FT4, free thyroxine; IQR, interquartile range; T3, total triiodothyronine; TSH, thyrotropin.

Disclosures

A.C.B. is a consultant for AbbVie, Acella, Synthonics, Thyron, and Madrigal. R.M.B.M. is an investigator of the Brazilian Research Council (CNPq). The other authors have nothing to disclose.

Data Availability

All data presented in this manuscript are contained within the figures and tables presented in the manuscript.

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