Thyroid Function and Body Weight: A Community-Based Longitudinal Study

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Published in:
PLOS ONE

Link to article, DOI:
10.1371/journal.pone.0093515

Publication date:
2014

Document Version
Publisher's PDF, also known as Version of record

Link back to DTU Orbit

Citation (APA):
Bjergved, L., Jørgensen, T., Perrild, H., Laurberg, P., Krejbjerg, A., Ovesen, L., ... Knudsen, N. (2014). Thyroid Function and Body Weight: A Community-Based Longitudinal Study. PLOS ONE, 9(4), [e93515]. DOI: 10.1371/journal.pone.0093515
Introduction

The thyroid gland produces thyroid hormones, which regulate gene transcription and metabolism throughout the body. Thyroid hormones play a key role in the basal metabolic rate, thermogenesis, as well as in the regulation of body metabolism in starved and fed states [1–3]. Overt thyroid disease is associated with marked changes in energy expenditure and body weight, with enhanced protein breakdown, lipolysis [4,5], and typically weight loss in hyperthyroidism, and the reverse in hypothyroidism [6]. Moreover, small changes in T4 dose in patients in long-term T4 treatment have been shown to modify resting energy expenditure significantly [7].

The main role of adipose tissues is to store energy to meet the energy needs of the body and to protect it from excess glucose by converting this to and storing it as triglycerides. Recently, adipose tissue has been recognized as a major endocrine organ, involved in the regulation of thermogenesis, food intake, and energy expenditure, e.g., the change between fed and starved state [3]. Studies of weight reduction by bariatric surgery [8] or gastric banding [9] have found postoperative changes in levels of TSH (serum thyrotropin), free T4 (thyroxine) and free T3 (triiodothyronine), but in different directions. Likewise, a decrease in TSH was observed following diet-, exercise-, and behavior therapy-induced weight loss in children and adolescents [10,11]. Conversely, obesity induced by high-fat diet weight gain has a significant effect on T4 and T3 levels in rats [12]. A relationship between body weight and thyroid hormones seems conceivable. The exact mechanisms are as yet unexplained and the temporality is debated.

Evidence from several cross-sectional population-based studies has shown a significant positive association between body mass index (BMI) and serum TSH, even for values of TSH within the normal reference range [13–18], among those a study by Knudsen et al. on baseline data from this cohort [16]. However, the number of longitudinal studies of the association are limited [14,17,19,20].
and the results are conflicting. The aim of the present study was to investigate the association between 11-year longitudinal changes in thyroid function as assessed by serum TSH and changes in body weight in a large sample of adult Danes. Furthermore, we wished to test the hypothesis that the relationship between serum TSH and free T4 is dependent on weight; a relation previous suggested to be a useful measure of the abnormal thyroid hormone sensitivity in a study of patients with hereditary pituitary resistance to thyroid hormones [21].

Materials and Methods

Ethics Statement

Participants gave their written informed consent, and studies were approved by the North Denmark Region Committee on Health Research Ethics (Nos. 2-16-4-0001-97, VN 96/208mch, and N-VN-19960208mch), and the Danish Data Protection Agency. The recommendations of the Declaration of Helsinki were followed.

Design and Population

The DanThyr program, described earlier in detail [22], is a Danish program monitoring the presence and development of thyroid disease before and after iodine fortification of household salt and salt used in the commercial production of bread since 1st July 2000. The present study used data from C1 cohort in the DanThyr program initiated in 1997/98 (C1a, N = 4,649, 50.1% participation) [23] and followed up in 2008–2010 (C1b, N = 2,465, 59.1% of baseline participants) [24]. The random samples were drawn from Copenhagen in the eastern part of Denmark and Aalborg in the western part of Denmark, representing areas with pre-fortification mild and moderate iodine deficiency, respectively. Using the national Civil Registration system, in which all inhabitants of Denmark can be identified by a unique 10 digit number, the sampling included four age groups of women (18–65 years) and one age group of men (60–65 years).

Participants underwent physical examination, gave a non-fasting venous blood sample and a spot urine sample, and had a thyroid ultrasonography (US) performed (Siemens Sonoline Versa Pro, Siemens, Erlagen, Germany). All procedures (e.g., invitation, questionnaires, and examination) were similar at baseline and follow-up examination. Blood and urine samples were collected between 08.00 a.m. and 5.30 p.m., stored at −20°C, and subsequently analyzed in random order with respect to region, sex, age, and season of the year at examination, at baseline, and follow-up. The normal reference range was defined in the baseline examination as: TSH, 0.4–3.6 mU/L; free T4, 9.8–20.4 pmol/L [23].

Among the 2,465 examined at both baseline and follow-up, 36 had missing serum TSH values, 28 had missing measurement of weight, 86 persons were pregnant, 228 had current or former treatment for thyroid disease (medicine, surgery, or radioactive iodine therapy), leaving 2,102 participants for analysis (Figure 1). Moreover, we replicated all analysis after exclusion of 282 participants with a baseline serum TSH concentration outside the normal reference range (below 0.4 mU/L or above 3.6 mU/L), leaving 1,944 participants for analysis. They were defined as having 1,944 participants for analysis. Moreover, we replicated all analysis after exclusion of 282 treatment for thyroid disease (medicine, surgery, or radioactive iodine therapy for thyroid disease, smoking status, and leisure time activity were self-reported. Smoking was classified as: never smokers, former smokers, occasional smokers, and daily smokers. According to the 11-year change in smoking status, participants were classified as: current smokers (N = 572), never smokers (N = 949), former smokers (N = 402), those who stopped smoking during follow-up (N = 381), and those who started smoking during follow-up (N = 98). A dichotomized smoking status (daily smoking, yes/no) was used to test the interaction with smoking in the three models. According to leisure time physical activity, participants were classified as mainly sedentary; light physical activity, 2–4 hours a week; light physical activity, more than 4 hours a week; or heavy physical activity, more than 4 hours a week. Eleven-year changes in physical activity were classified as less active, unchanged, or more active.

Statistical Analysis

Data were analyzed using SAS, version 9.2 (SAS Institute Inc. Cary, NC, USA). Two-sided probability values <0.05 were considered significant. Non-normally distributed variables were log transformed to obtain a normal distribution. Descriptive characteristics of the participants presented as percent (total) or median (interquartile range, IQR) were compared with non-parametric statistics, Kruskal-Wallis test. We used multiple linear regression analysis to model the associations in the following three models: 1) the association between baseline serum TSH and change in weight during follow-up; 2) the association between change in serum TSH and change in weight during follow-up; and 3) the association between baseline BMI and change in serum TSH concentration during follow-up. Furthermore, we modeled linear regressions of: 1) the association between baseline free T4 and change in weight during follow-up; and 2) the association between change in free T4 and change in weight. Men and
women were analyzed separately because of the age and sex composition of the population. All the models were adjusted for the baseline value of the outcome to adjust for regression to the mean and to allow changes in the exposure variables to be interpreted as effects on outcome changes from baseline to follow-up [29]. In addition, we adjusted for age at baseline (categorical), change in smoking status, and change in leisure time physical activity. Because the age range was only 5 years for men, the age variable was left out in their analysis. We also divided the exposure variables in the above described regression models in quartiles to test for a non-linear relationship between predictor and outcome.

In additional analysis, the relationship between Δ TSH (TSH concentration at follow-up minus TSH concentration at baseline, as outcome) and Δ free T4 concentration (as predictor) in three weight change groups (tertiles) was evaluated by linear regression analysis, adjusting for age group, change in smoking, and change in leisure time physical activity (and for the baseline value of serum TSH). We added an interaction term between Δ free T4 and

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**Figure 1. Flow chart of the study group.** TSH serum thyrotropin. doi:10.1371/journal.pone.0093515.g001
weight change tertiles to test if the regression lines for the three weight groups were significantly different. Slopes of the three regression lines were identified from the same model. This analysis was restricted to women.

We tested the validity of all linear models by checking normality of residuals, homogeneity of residuals, and linearity of the relationship between response variable and predictors.

Wilcoxon Rank test was used to compare the independent measures of baseline characteristics of participants and non-participants at the re-examination.

**Results**

Descriptive characteristics of the participants are shown in Table 1. The average follow-up time was 11.2 years (range, 10.1–12.8 years). Weight increased by 3.0 kg (95% CI, 2.7–3.3 kg) during follow-up; weight did not change significantly in men, 60–65 years at baseline (−0.2 kg, 95% CI, −0.9–0.4 kg), whereas it increased significantly by 3.7 kg (95% CI, 3.4–4.1 kg) in women (18–65 years at baseline). The median serum TSH increased significantly from the first to the second examination in the total population of individuals (from 1.30 (95% CI, 0.91–1.89) to 1.44 (95% CI, 0.99–2.05), P < 0.01) and among men (from 1.24 (95% CI, 0.83–1.82) to 1.40 (95% CI, 0.94–2.02), P < 0.01) as well as among women (from 1.31 (95% CI, 0.92–1.90) to 1.45 (95% CI 1.00–2.06), P < 0.01).

The median serum TSH increased significantly from the first to the second examination in the total population of individuals (P < 0.01), as well as among men (P < 0.01) and women (P < 0.01).

In the adjusted model, we found no association between baseline log TSH and weight change during follow-up in either sex among individuals without treatment for thyroid disease (women, P = 0.17; men, P = 0.72) and in persons with a serum TSH within the normal reference range at baseline (women, P = 0.19; men, P = 0.06). No association between baseline log BMI (modeled as a continuous variable) and TSH change was found, neither in all non-treated persons (women, P = 0.21; men, P = 0.83) nor in those with a baseline TSH concentration within the normal reference range (women, P = 0.78; men, P = 0.69).

A significant association between change in serum TSH concentrations and weight change was observed in both sexes (Table 2). In the adjusted model, weight increased by 0.3 kg (95% CI, 0.1–0.4) for every one-unit increment in serum TSH (mU/L) (P = 0.005) in women free of thyroid disease. In men, a one-unit increment in TSH was associated with a 0.8 kg (95% CI, 0.1–0.4) for every one-unit increment in serum TSH among women, and by 0.7 kg (95% CI, 0.02–1.3, P = 0.04) for every unit increment in TSH among men. Women with a serum TSH change in the lowest quartile had a mean weight increase of 1.2 kg (IQR −2.8, 5.4), whereas women

| Characteristics | All participants % (N) | Median (IQR) | Follow-up |
|-----------------|------------------------|--------------|-----------|
|                 | Median (IQR)           | Serum TSH (mU/L) | Median (IQR) | Serum TSH (mU/L) | BMI (kg/m²) | BMI (kg/m²) |
| Age (years)     |                        |               |            |               |            |            |
| 18–20, women    | 19.7% (415)            | 1.42 (0.98, 1.98) | 22.1 (20.4, 24.6) | 1.46 (1.11, 2.00) | 23.6 (21.2, 26.9) |            |
| 25–30, women    | 20.8% (436)            | 1.36 (0.99, 1.92) | 22.4 (20.6, 24.3) | 1.49 (1.04, 2.09) | 23.5 (21.5, 26.6) |            |
| 40–45, women    | 26.9% (566)            | 1.23 (0.85, 1.78) | 24.1 (21.9, 27.0) | 1.42 (0.93, 2.07) | 25.5 (23.0, 28.9) |            |
| 60–65, women    | 13.6% (286)            | 1.31 (0.84, 1.96) | 26.2 (23.9, 29.0) | 1.40 (0.94, 2.11) | 26.6 (24.1, 29.7) |            |
| 60–65, men      | 19.0% (399)            | 1.24 (0.83, 1.82) | 26.3 (24.6, 28.9) | 1.40 (0.94, 2.02) | 26.6 (24.9, 29.5) |            |
| P values*       |                        | 0.0019        | <0.0001    | 0.39          | <0.0001       |            |
| Leisure time physical activity | | | | | | |
| Sedentary       | 11.4% (237)            | 1.22 (0.91, 1.92) | 23.9 (21.5, 28.1) | 1.40 (0.94, 2.09) | 26.7 (22.9, 30.9) |            |
| Light           | 45.9% (956)            | 1.28 (0.90, 1.82) | 24.5 (21.9, 27.4) | 1.41 (0.98, 2.00) | 25.5 (22.8, 28.9) |            |
| Moderate/vigorous | 42.7% (889)         | 1.33 (0.92, 1.92) | 23.9 (21.7, 26.3) | 1.49 (1.02, 2.07) | 24.8 (22.3, 27.3) |            |
| P value*        |                        | 0.34          | 0.009      | 0.24          | <0.0001       |            |
| Smoking status  |                        |               |            |               |            |            |
| Current smoker  | 32.3% (678)            | 1.21 (0.84, 1.76) | 23.5 (21.2, 26.3) | 1.22 (0.85, 1.78) | 24.8 (22.1, 28.1) |            |
| Never smoker    | 40.2% (846)            | 1.35 (0.96, 1.99) | 24.0 (21.4, 26.8) | 1.56 (1.10, 2.16) | 25.1 (22.4, 28.2) |            |
| Former smoker   | 19.6% (413)            | 1.30 (0.88, 1.97) | 25.5 (23.2, 28.2) | 1.43 (0.98, 2.08) | 25.7 (23.3, 29.0) |            |
| Occasional smoker | 7.8% (165)           | 1.28 (0.82, 1.84) | 23.8 (22.1, 27.0) | 1.43 (1.00, 1.98) | 24.7 (22.3, 28.4) |            |
| P value*        |                        | 0.0003        | <0.0001    | <0.0001       | 0.0002        |            |
| Region          |                        |               |            |               |            |            |
| Copenhagen     | 50.7% (1,065)          | 1.34 (0.92, 1.91) | 24.1 (21.7, 27.1) | 1.53 (1.06, 2.17) | 25.0 (22.5, 28.2) |            |
| Aalborg        | 49.3% (1,037)          | 1.27 (0.87, 1.82) | 24.1 (21.8, 26.8) | 1.35 (0.90, 1.92) | 25.5 (22.7, 28.6) |            |
| P value*       |                        | 0.03          | 0.68       | <0.0001       | 0.11          |            |
| TPO-Ab status (kU/L) |            |               |            |               |            |            |
| ≥30            | 21.1% (443)            | 1.42 (0.96, 2.12) | 23.5 (21.7, 26.7) | 1.78 (1.15, 2.80) | 24.9 (22.8, 28.1) |            |
| <30            | 78.9% (1,655)          | 1.28 (0.89, 1.82) | 24.3 (21.8, 27.1) | 1.39 (0.96, 1.91) | 25.3 (22.6, 28.5) |            |
| P value*       |                        | 0.0001        | 0.057      | <0.0001       | 0.52          |            |

*Kruskal-Wallis test.

TSH serum thyrotropin, BMI body mass index, TPO-Ab thyroid peroxidase antibody, CI confidence interval, IQR interquartile range.

doi:10.1371/journal.pone.0093515.t001

Table 1. Serum thyrotropin status and body mass index according to characteristics.
Among the 4,649 persons participating in the DanThyr baseline study, 405 persons died and 72 emigrated before or during the 11-year follow-up. Comparison of these persons with the survivors who participated in the follow-up showed no difference between the groups concerning baseline values of serum TSH concentrations (P = 0.40), weight (P = 0.75), and BMI (P = 0.08), as previously reported [24].

Discussion

In this community-based sample of 1,944 adults followed for 11 years, we found a statistically significant positive association between change in serum TSH concentrations over time and weight change. However, baseline TSH concentration was not a determinant of change in weight and baseline BMI level was not a determinant of change in TSH during follow-up. The relationship between change in TSH and free T4 change differed significantly between groups with different weight changes.

Several cross-sectional population studies found a significant positive association between serum TSH levels and BMI or body weight in individuals free of thyroid disease, in both sexes [13,16], or in women only [15]. Also, in persons with TSH levels within the normal reference range, a significant association with body weight was confirmed [14,18], though restricted to non-smoking subjects in one study [17]. Two studies could not confirm the association [30,31]. Possible explanations for the lack of association found by Manji et al. [31] could be the small sample size (N = 400), the different clinical spectrum of the studied population (euthyroid hospital-referred patients with thyroid nodules or goiter), and the use of correlations without adjustment for confounders. Our result regarding the significant, positive association between change in serum TSH and change in body weight is in line with the results of the four existing longitudinal population studies [14,17,19,20]. However, the association was restricted to non-smokers in the Tromsø study [17]. Regarding the observed lack of association between baseline TSH and weight change during follow-up, the same was observed by Fox et al. [14] and Svare et al. [20]. Svare et al. [20], contrary to our study, found a negative association between baseline BMI and change in serum TSH. Fox et al. [14] and Nyrnes et al. [17] did not investigate the association between baseline BMI and TSH change.
Fat tissue could affect thyroid hormones in a number of ways. First, leptin, an adipocyte-derived hormone, could affect thyroid hormone status through the stimulation of hypothalamic thyrotropin-releasing hormone (TRH) gene expression [32,33] and influence thermogenesis [9]. However, as described in a recent review, the role of leptin seems to be restricted to energy-deficient states such as acute fasting and exercise, and evidence in humans is still scarce [34]. Second, the effect may be due to a change in deiodinase activity in central and/or peripheral tissue [2,9]. Third, hormone resistance in obese subjects as a result of a T3 receptor decrease has been suggested [35]. Fourth, a partially bioinactive TSH protein in obese subjects has been speculated [33].

Our longitudinal results provide insight into the association between body weight and TSH found in cross-sectional observational studies. An association between weight and serum TSH seems to be present at all time points. However, the association we found does not necessarily reflect a causal relationship. Both TSH and weight might be modified by a third factor. The absence of a predictive value of TSH for weight change and of BMI for TSH change could suggest that this might be the case.

A modifying effect of tobacco smoking on the relationship between thyroid function and BMI in euthyroid subjects has been suggested [30]. Our analyses do not, however, support tobacco smoking as a modifying factor, as we found no significant interaction with change in smoking status in any of the models.

Previous studies of the TSH/free T4 relationship have shown an exponential decrease in TSH with progressive increase in serum T4 [35,36]. Ercan-Fang et al. suggested [21], in a study of patients with hereditary pituitary resistance to thyroid hormones, that assessment of the slope of lnTSH as a function of free T4 was a useful measure of the abnormal thyroid hormone sensitivity. The exact mechanism behind the observed difference in the ΔTSH/Δ free T4-relation for the upper, middle and lower tertiles of weight change, can not be resolved on the basis of this study. Possible suggestions could however be a higher pituitary response to change in free T4 with increasing weight, a less biological active TSH molecule with increasing weight, or even more speculative, that the thyroid glands response is less sensitive to TSH with increasing weight.

Our cohort was exposed to a program of mandatory iodization of salt, resulting in an increased iodine urine excretion [24]. It may be speculated that the increased iodine intake could affect the studied association. To the best of our knowledge, no studies have described an association between iodine intake and weight, which speaks against iodine as a possible confounder of the studied associations. Moreover, the absence of interaction with region (as a proxy for different levels of iodine intake) in the adjusted model of weight change and TSH change and the fact that creatinine-adjusted iodine excretion was not associated with change in weight imply that the iodization program was unlikely to have had a major effect on the studied association.

The strengths of our study are the longitudinal population-based design, ensuring the temporality of studied associations; the long-term follow-up; the objective measurement of exposures and outcomes; the measurement of free T4 beside TSH at baseline and follow-up; and the detailed information on covariates. The limitations could be residual confounding from unmeasured confounders such as diet or non-thyroidal illness; and selection bias as a consequence of the relatively low participation rate of 59.1%. Furthermore, TSH is a measurement known to have a large within individual variation and analytical variation [37], and the fact that the determination of TSH relied on a single measurement at baseline and at follow-up could bias the values of change in TSH during follow-up. This would, however, only lead to a non-differential misclassification. Repeated measurements of TSH and weight in each individual during the follow-up period would more precisely outline the shape of their relationship with time [30].
Conclusion

In conclusion, TSH levels are not a determinant of future weight changes and BMI is not a determinant of TSH changes, but changes in TSH levels seem to be positively associated with weight change. It remains to be proven whether body weight and serum TSH are causally connected.

Acknowledgments

We are indebted to laboratory technicians Ingelise Leegard and René Fiege for invaluable assistance with thyroid ultrasound investigations and biochemical analysis, to MD Mathilde Svendstrup for assistance with data collection, to Carsten Agger for support in data management, and to all the participants of the study for their important contribution.

Author Contributions

Conceived and designed the experiments: TJ HP PL LO LBR NK. Analyzed the data: LB TJ NK. Contributed reagents/materials/analysis tools: NK AK LB. Wrote the paper: LB. Revised the manuscript critically and approved the final manuscript for publication: LB TJ HP PL LO LBR NK.

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