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Serum leptin, thyroxine and thyroid stimulating hormone levels interact to affect cognitive function among US adults: evidence from a large representative survey

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Abstract

Neuroanatomical connections point to possible interactions between areas influencing energy homeostasis and those influencing cognition. We assessed whether serum leptin, thyroxine and thyroid stimulating hormone (TSH) levels are associated with and interact to influence cognitive performance among US adults. Data from the National Health and Nutrition Examination Survey III (1988–94) were used. Measures included a battery of neuropsychological tests and serum leptin, thyroxine and TSH levels (20–59yo: n=1114–2665; 60–90yo: n=1365–5519). Among those 20–59yo, the middle tertile of leptin (*vs.* first tertile) was inversely related to the number of errors on the symbol digits substitution test. Increased thyroxine level was associated with a poorer performance on the serial digits test in 20–59yo, but a better performance on the Math test in 60–90yo. TSH was associated with poor performance on various tests in 20–59yo, but better performance in 60–90yo. Significant antagonistic interactions were found in both age groups between thyroxine, TSH and leptin for a number of tests, including between leptin and thyroxine in 60–90yo in their association with word recall-correct score. We found significant associations of our main exposures with cognitive function among US adults, going in opposite directions between age groups in the cases of thyroid hormonal levels, as well as some interactive effects between exposures. It is important to conduct prospective cohort studies to provide further insight into potential interventions that would assess interactive effects of various hormonal replacement regimens.

Keywords

Leptin; thyroxine; thyroid stimulating hormone; cognitive function; aging

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INTRODUCTION

Thyroid hormones and the adipokine hormone leptin are both determinants of adiposity, through the regulation of energy metabolism, thermogenesis, glucose and lipid metabolism, appetite and food intake, and the oxidation of fatty acids (Biondi, 2010, Farooqi, et al., 2007, Jacob, et al., 1997, Zhang, et al., 1994). Findings from a recent meta-analysis (Beydoun, et al., 2008) linked poor cognitive function, and increased Alzheimer's Disease risk, by approximately 80% to obesity (i.e. higher adiposity status as measured by body mass index 30 kg.m^{-2}). Hormonal mechanisms underlying the association between obesity and poor cognition are still understudied. Interestingly, thyroid hormones are also key actors on the central nervous system during neuro-development (Anderson, 2001, Dugbartey, 1998), by regulating nervous system myelination, growth, puberty, metabolism and organ functions (Smith, et al., 2002). In adults (Hendrick, et al., 1998, Loosen, 1992), the brain becomes more sensitive to thyroid function changes, a rationale behind using thyroid hormones to treat psychiatric and affective disorders (Bauer and Whybrow, 2001). Additionally, hypothyroidism was linked to progressive cognitive impairment and slower thought processes, leading to pseudo-dementia, a condition used to rule out primary degenerative dementia (Dugbartey, 1998, Loosen, 1992). However, despite the well-established effects of hypothyroidism on cognitive function, the effects of hormonal levels within normal ranges are less well-studied. Research suggests that thyroxine (T_4) and thyroid stimulating hormone (TSH) fluctuations within normal or pre-clinical ranges may be associated with cognitive performance. Evidence suggests hypothyroidism as a potential risk factor for cognitive impairment in some studies (Bono, et al., 2004, Burmeister, et al., 2001, Correia, et al., 2009, Mafrica and Fodale, 2008, Miller, et al., 2006, Monzani, et al., 1993, Osterweil, et al., 1992, Prinz, et al., 1999, Samuels, et al., 2007a, Volpato, et al., 2002, Wahlin, et al., 1998) but not others (Almeida, et al., 2007, Kramer, et al., 2009, Samuels, et al., 2007b).

New evidence suggests that the adipokine hormone leptin, originally linked to appetite and eating behavior by acting on the hypothalamus and striatal brain regions (Farooqi, et al., 2007, Jacob, et al., 1997, Zhang, et al., 1994), has pleiotropic effects within the cortex and hippocampus involved in cognition. In fact, when leptin was administered directly into the hippocampus of mice, it was shown to improve memory processing and to shape the hypothalamus during the earliest stages (Harvey, et al., 2005). A number of recent epidemiological studies indicate an association (Gunstad, et al., 2008, Holden, et al., 2009, Paz-Filho, et al., 2008), yet it remains unclear whether there are interactions between areas influencing energy homeostasis and cognition, despite well-established neuroanatomical connections suggesting this (Morrison, 2009). As both thyroid hormones and leptin may be involved in cognition by potentially mediating the effect of adiposity on brain function, it is important to assess whether their effects are interactive or simply additive. The present study used cross-sectional nationally representative data from the National Health and Nutrition Examination Survey (NHANES) III to examine associations and interactions of leptin, T_4 and TSH levels with various tests of cognitive performance among adults in the United States.

METHODS

Participants

NHANES III (1988–1994), a multistage, stratified sampling design, over-sampled older adults (60–90yo) and minorities (African Americans and Mexican Americans). Following a home-interview, selected individuals were invited to a mobile examination center (MEC) for body measurements, clinical evaluations and laboratory testing (Center for Disease Control and Prevention (CDC), 1996). Among adults (aged 20–90yo) interviewed, 12,264 were 20–59yo and 6,033 were 60–90yo. Two separate and distinctive batteries of cognitive function

tests were administered to those two age groups, respectively. Cognitive testing was available on a smaller proportion (n=3288 of 12,264) of 20–59yo compared to 60–90yo (n=5811 to 6033 out of 6,033). Among participants with complete cognitive, demographic, lifestyle and health-related data, thyroxine and TSH data were generally complete in both age groups (2,496 to 2,665: 20–59yo and 4,086 to 5,519: 60–90yo). However, a small proportion of those with complete cognitive data also had leptin data (1,114 to 1,164: 20–59yo; 1,365 to 1,420: 60–90yo) (See Figure 1).

Cognitive assessment

As part of the MEC exam, a battery of three cognitive tests was administered to participants aged 20–59yo and considered in our present study: simple reaction time (**SRT**), symbol-digit substitution test (**SDS**) and serial digits learning (**SDL**), from which five cognitive test scores were computed. Among those aged 60–90yo, three cognitive tests were administered and used in our present study: Word recall (**WR**), story recall (**SR**), a math test (also termed serial 3' s; **MATH**) and orientation to time from the mini-mental state examination (**ORIENT**), (Folstein, 1975) (See Online Supplemental Material).

Laboratory assessments

Serum leptin—After an overnight fast, surplus sera from 6,415 participants (20–90yo) was tested for leptin level (in Fg/L) by the laboratory Linco Research, Inc., St. Louis, Missouri, using a radioimmunoassay (RIA) with a polyclonal antibody raised in rabbits against highly purified recombinant human leptin. The minimum detectable concentration of the assay was 0.5 Fg/L leptin and the limit of linearity was 100 Fg/L. Recovery of leptin added to serum is 99–104% over the linear range of the assay. The RIA agrees reasonably well with rough quantification by Western blot. Within-and between-assay CVs range from 3.4% to 8.3% and from 3.6% to 6.2%, respectively. (US DHHS, 2002)

Serum total circulating thyroxine (T₄)—Thyroxine (in µg/dL; 1 nmol/L=12.8 µg/dL) testing was performed on examinees aged 13–90yo. Total (protein-bound and free) circulating thyroxine (T₄) concentrations were determined using an enzyme-based homogeneous immunoassay on the Hitachi 704. The reference range is 4.5 to 12.5 µg/dL. The coefficient for inter-assay variation (CV) was less than 10%. (Gunter, 2010)

Serum thyroid stimulating hormone—The thyroid stimulating hormone (TSH, in mu/L) assay is a chemiluminescence immunometric assay utilizing a mouse monoclonal antibody to TSH immobilized on a polystyrene bead and a goat polyclonal antibody to TSH conjugated with an acridinium ester. The reference range is 0.3 to 5.0 mu/L. The coefficient for inter-assay variation (CV) was less than 5%. (Gunter, 2010)

Covariates

In multiple age group-specific (20–59yo; 60–90yo) regression models, covariates considered as potential confounders included age (continuous), sex, race/ethnicity (Non-Hispanic White (NHW), Non-Hispanic black (NHB), Mexican-American (MA), Other ethnicity (OTHER)), marital status (0=unmarried; 1=currently married), educational level (<High school, High school, >High school), poverty income ratio (100%; >100- 200%; >200%), language of interview (1=English; 2=Spanish) and smoking status coded as: “Never”, “Former” and “Current”. Moreover, self-reported physical activity compared to age peers was selected, with possible responses: “More active”; “About the same” and “less active”. Weight status was measured using the body mass index (weight(kg) divided by squared height (m⁻²)).

To assess overall dietary quality, a widely-used and readily available diet quality index, the USDA's 1995-Health Eating Index (HEI) (McCullough, et al., 2000) (range: 0–100) was employed, with higher scores reflecting better dietary quality. Quintiles score were computed and entered into models as four dummy variables (common referent: lowest quintile).

Participants self-rated their health as follows: Would you say your health in general is "Excellent", "Very good", "Good", "Fair" or "Poor"? The variable was entered into models as four dummy variables (common referent: "Excellent"). Finally, an index was created using self-reported co-morbid conditions namely "arthritis", "congestive heart failure", "stroke", "asthma", "chronic bronchitis", "emphysema", "hay fever", "cataracts", "goiter", "thyroid disease", "lupus", "gout", "skin cancer", "other cancer", "diabetes", "hypertension", "chest pain/possible angina", "heart attack/myocardial infarction", "osteoporosis" and "kidney stones". Thus, a participant's value on this index may range between 0 and 20.

Data analysis

Stata release 11.0 (STATA, 2009) was used for analyzing survey data. Two design variables were specified for the six-year period of NHANES III, namely the primary sampling unit and the stratum. Sampling weights used were those for cognitive testing variables (six-year) in the case of 20–59yo sub-group and those for the MEC exam (six-year) for those aged 60–90yo. Means, proportions and regression coefficients were estimated taking into account design complexity and sampling weights. Main exposures were Log_e transformed and categorized as tertiles in the major part of the analysis, which consisted of the following steps: First, means of Log_e transformed leptin, thyroxine and TSH were graphically represented using dot plots, comparing age group-sex categories in one analysis and age group-race in another. To statistically test differences between categories in each analysis and for each exposure variable, ANOVA test was conducted taking sampling weights into account, followed by bonferroni-corrected multiple comparisons. Second, sample characteristics were described, compared between the two age groups (by assessing 95% CI overlap) and then entered into a multiple OLS regression model as predictors for the three main exposure variables (Log_e transformed). Third, for each age subgroup, multiple OLS and zero-inflated poisson regression models were conducted with cognitive function scores as outcomes and the three main exposure variables (Log_e transformed and converted to tertiles) entered separately in each model along with potentially confounding socio-demographic, lifestyle and health-related factors.

OLS models were conducted for continuous pseudo-normal cognitive scores (usually involving time elapsed to complete the test; 20–59yo), whereas zero-inflated poisson regression models were conducted in the tests for 60–90yo which were count outcomes with a large proportion scoring as zero (e.g. no errors on the test=zero score). Finally, pairwise interaction between the three main exposures was tested in OLS and the zero-inflated poisson models. Additionally, three secondary analyses were conducted: First, effect modification by sex and race of the associations between the three exposures and the various cognitive outcomes was tested by adding interaction terms alternatively with sex and race to the main effects model. Second, analyses were repeated for the total sample after excluding participants with extreme values on thyroxine and TSH (i.e. outside the reference range). A type I error of 0.05 was considered for statistical significance in all analyses, except for interaction terms where significance level was set at 0.10 (Selvin, 2004).

RESULTS

Study sample characteristics

Table 1 displays estimated means and proportions of main study characteristics. Generally, the younger age group (20–59yo) had a higher proportion of participants using Spanish as the interview language (5.2% vs. 1.4%), more current smokers (33.5% vs. 18.7%), excellent self-rated health (23.4% vs. 10.2%) and were more active than age peers (22.0% vs. 13.5%). However, the older age group (60–90yo) had an overall healthier diet (1995-HEI: 65.7 vs. 61.8), but a higher mean co-morbidity index (2.6 vs. 1.07). Leptin, thyroxine and TSH levels were significantly elevated in the 60–90yo group.

Associations of leptin, thyroxine and thyroid stimulating hormone with socio-demographic, lifestyle and health-related factors

Based on Figure 2 which examined and compared weighted means of each Log_e transformed exposure across age group-sex categories, the following patterns were observed within age group and across sex: First, leptin and T_4 levels were significantly more elevated among women compared to men in both age groups, while TSH level was significantly higher among women in the 60–90yo group but lower among women in the 20–59yo age group, compared to men in that age group. Figure 3 shows significant race differences in levels of Log_e transformed exposures within each age group. For instance, leptin level was significantly lower among NHW particularly within the 20–59yo age group (compared to NHB and MA). T_4 level on the other hand did not differ significantly within the 60–90yo age group and across race, while in the 20–59yo group, NHW had significantly lower levels compared to MA and OTH 20–59yo. TSH level was significantly lower within the 20–59yo group among NHB compared to NHW and the other two race groups. In contrast, in the 60–90yo group, NHB had the highest TSH level compared to the other three race groups.

Based on findings from OLS regression analyses (Table 2), leptin was significantly higher in women in both age groups. It was positively associated with age in the 20–59yo group, but inversely related to age in the 60–90yo group. In the 20–59yo group, leptin was significantly higher among NHB and MA (compared to NHW) and lower among current cigarette smokers (compared to never smokers). It showed an increasing trend with poorer self-rated health, and a decreasing trend with lower physical activity and higher co-morbidity index (20–59yo group) and more importantly a positive association with BMI in both age groups. Thyroxine (20–59yo group) was significantly higher among NHB and lower in OTHER compared to NHW. In the 60–90yo group, thyroxine was positively associated with the 1995-HEI and inversely related to BMI. TSH, on the other hand, was positively associated with age and inversely related to smoking status in the 20–59yo group and it was inversely related to educational attainment and BMI in the 60–90yo group.

The prevalence of hyperthyroidism (i.e. reference range exceeded values) in 20–59yo was 4% for thyroxine and 2.5% for TSH. In 60–90yo prevalence rates were 3% and 7%, respectively. In terms of hypothyroidism (i.e. below the reference range), prevalence was 1.7% for thyroxine and 2.1% for TSH in 20–59yo. Rates were 2.9% and 6.6%, respectively for 60–90yo.

Associations of leptin, thyroxine and thyroid stimulating hormone with cognitive performance in the 20–59yo and 60–90yo age groups: Multiple OLS and zero-inflated poisson regression models

Table 3 displays associations between the three main exposures and cognitive performance, based on multiple regression analyses. The middle tertile of leptin (vs. first tertile) was inversely related to number of errors on the symbol digits substitution test, indicating better

cognition at that level (**SDS-E**) in 20–59yo (Table 3, **Model 1**). A secondary analysis examined effect modification by race. This association was stronger among NHW compared to NHB and MA ($p < 0.05$ for associated interaction terms).

The more elevated thyroxine level was (Table 3, **Model 2**), the more the worse the score was on serial digits test, trials to completion (**SDL-TTC**) in 20–59yo, when comparing the second to the first tertile. However, this association became only borderline significant when restricting thyroxine and TSH levels to the reference range ($p < 0.10$). Conversely, thyroxine was associated with better performance on the Math test (lower **MATH-INC**) and the story recall test (higher **SR-CORR**) in 60–90yo (Table 3, **Model 2**), findings that were not altered by restricting the sample to participants within the reference ranges of thyroxine and TSH (data not shown).

TSH was associated with a number of cognitive performance scores for both age groups. In 20–59yo, a higher TSH was linked to poorer performance on the simple reaction time and symbol digits substitution test-latencies (higher **SRT** and **SDS-L**), though the association with **SRT** became non-significant when participants outside the reference ranges of thyroxine and TSH were excluded (data not shown). In contrast, increased serum level of TSH for 60–90 yo was associated with improved performance on Word recall-correct, story recall-correct, and on the Math and orientation tests. In two tests (**SR-CORR** and **MATH-INC**), the association was significantly stronger among men ($p < 0.05$ for TSH $\times$ sex interaction term). However, only the association between TSH and **SR-CORR** (third vs. first tertile) and between TSH and **MATH-INC** (second vs. first tertile) remained statistically significant when participants outside the reference ranges of thyroxine and TSH were excluded (data not shown).

Interactions between leptin, thyroxine and TSH in their association with cognitive performance in the 20–59yo and 60–90yo age groups: Multiple OLS and zero-inflated poisson regression models

In Table 4, we tested interaction between exposures (two at a time) in affecting cognitive performance. In **model 1** (with thyroxine, TSH and thyroxine $\times$ TSH), a significant antagonistic interaction (i.e. sign of the interaction term was opposite to that of the main effects) was found in both age groups, for the following cognitive performance tasks: **SDS-L**, **SDL-TE**, **SR-CORR**, and **MATH-INC** ($P < 0.05$ for thyroxine $\times$ TSH interaction term). In **model 2**, an antagonistic interaction of thyroxine and leptin was observed for **WR-CORR**, whereby a potentially beneficial effect of thyroxine on that cognitive score was stronger at the lowest tertile of leptin ($p < 0.05$ for leptin $\times$ thyroxine interaction term that had a positive sign in contrast to main effects of thyroxine and leptin, which had a negative sign).

Finally, there were also antagonistic interactions between TSH and leptin (**model 3**) in their associations with **SDL-TE** and **SDL-TTC**. In particular, higher levels of TSH were associated with better cognition, particularly at lower levels of leptin. Most significant findings were unaltered by excluding participants outside the reference ranges of thyroxine and TSH. (data not shown).

DISCUSSION

This cross-sectional study of a nationally representative sample of US adults examined the associations of leptin, thyroxine and TSH with cognitive test performance in 20–59yo and 60–90yo. We also examined interactions between the three exposures in their associations with cognitive performance. Among those 20–59yo, the middle tertile of leptin (vs. first tertile) was inversely related to the number of errors on the symbol digits substitution test. Increased thyroxine level was associated with a poorer performance on the serial digits test

in 20–59yo, but a better performance on the Math and story recall tests in 60–90yo. TSH was associated with poor performance on various tests in 20–59yo, but better performance in 60–90yo. Significant antagonistic interactions were found in both age groups between thyroxine, TSH and leptin for a number of tests, including between leptin and thyroxine in 60–90yo in their association with word recall-correct score as well as leptin and TSH in 20–59yo in their association with serial digits learning test scores.

Overall, a limited number of cross-sectional, cohort and experimental studies have examined the effect of thyroid function on various cognitive outcomes. Out of ten cross-sectional studies (Almeida, et al., 2007, Cardenas-Ibarra, et al., 2008, Ceresini, et al., 2009, Kramer, et al., 2009, Prinz, et al., 1999, Samuels, et al., 2007b, Stern, et al., 2004, Stuerenburg, et al., 2006, Wahlin, et al., 1998, Wu, et al., 2006), six suggested that either hypothyroidism or hyperthyroidism may be linked to adverse cognitive outcomes, two had mixed results (Wahlin, et al., 1998, Wu, et al., 2006) and two had no significant results (Almeida, et al., 2007, Kramer, et al., 2009). Out of four cohort studies (de Jong, et al., 2009, Gussekloo, et al., 2004, Hogervorst, et al., 2008, Volpato, et al., 2002), three suggested positive (de Jong, et al., 2009, Hogervorst, et al., 2008, Volpato, et al., 2002), and one suggested negative findings (Gussekloo, et al., 2004). In one of the positive studies (de Jong, et al., 2009), 1 standard deviation higher serum free thyroxine was associated on average with 20–30% increased risk of dementia and AD among Japanese-American men aged 71–93y (n=665). Another large cohort study (n=599) conducted in the Netherlands, also among a group of older adults, did not find an association between thyroid status at baseline and cognitive function (Gussekloo, et al., 2004). Out of seven experimental studies (Bono, et al., 2004, Burmeister, et al., 2001, Correia, et al., 2009, Miller, et al., 2006, Monzani, et al., 1993, Osterweil, et al., 1992, Samuels, et al., 2007b), four had positive findings (Bono, et al., 2004, Correia, et al., 2009, Monzani, et al., 1993, Samuels, et al., 2007b) and three had mixed findings (Burmeister, et al., 2001, Miller, et al., 2006, Osterweil, et al., 1992). For instance, in an intervention study on 21 hypothyroids and 17 sub-clinical hypothyroid patients, neuropsychological testing was administered at baseline and then 3 and 6 months after T₄ replacement and compared to normal participants. After T₄ replacement, verbal memory normalized in both overt and sub-clinical hypothyroid groups. Spatial memory normalized in the sub-clinical group but remained impaired in the overtly hypothyroid group. Associative memory deficits persisted in the overt hypothyroid group (Correia, et al., 2009).

Our findings are of great importance as they point to different directions of associations between thyroid hormones and cognition in younger versus older adults. In particular, among younger and middle-aged adults, thyroxine (T₄) and TSH were shown to have a deleterious effect on cognition in the domains of learning, reaction time and psychomotor speed. Few studies have investigated the same domains and population, but some similar findings have been observed in earlier studies. For instance, an investigation of adult hypothyroid patients demonstrated deficits in the areas of verbal learning and psychomotor functioning, that was partially ameliorated with thyroid treatment (Osterweil, et al., 1992). In contrast, among older adults, thyroxine and TSH were shown to be related to an improved cognitive performance in domains of verbal memory, attention/calculation and orientation to time. Similar findings were observed in a number of previous cross-sectional (Ceresini, et al., 2009, Kramer, et al., 2009) and longitudinal studies (Miller, et al., 2006, Samuels, et al., 2007a, Volpato, et al., 2002). For instance, older adults with subclinical hypothyroidism had poorer MMSE scores (Kramer, et al., 2009) than euthyroid adults, and MMSE scores for adults who underwent 20 years of thyroid treatment were no different than euthyroid adults (Miller, et al., 2006). Longitudinally, hypothyroid patients who underwent 3 months of thyroxine treatment increased their performance on verbal memory (Miller, et al., 2006), while older women with greater 3-year thyroxine declines (lowest thyroxine tertile) had

greater decreases in MMSE score (Volpato, et al., 2002) when compared with women in the highest thyroxine tertile.

We identified one cross-sectional (Gunstad, et al., 2008) and two cohort studies (Holden, et al., 2009, Lieb, et al., 2009) that examined the effect of leptin levels on cognition, dementia or Alzheimer's disease. In the cohort study (Health ABC, n=2871, age: 70–79y), participants in the high leptin group were at 34% lower risk for clinically significant cognitive decline over 4 years (5-point drop on the 3MS) compared to the lowest group; (OR=0.66; (95% CI: 0.48–0.91), independent of co-morbidities and body fat (Holden, et al., 2009). In another more recent prospective study of 785 persons without dementia (Framingham study, mean age (SD), 79y (5) 62% female), higher leptin was associated with lower risk of incident dementia and AD in multivariable models (hazard ratio per 1-SD increment in log leptin was 0.68 (95% confidence interval, 0.54–0.87) for all-cause dementia and 0.60 (95% confidence interval, 0.46–0.79) for AD (Lieb, et al., 2009). In contrast, in the cross-sectional study (n=35; mean age: 74y), serum leptin was inversely related to speeded executive function in older adults who had no significant neurological or psychiatric conditions (Gunstad, et al., 2008).

Various biological mechanisms can explain the potential associations between thyroxine, TSH and cognition. First, the concentrations of both T_4 and its more potent metabolite T_3 are preserved at narrow ranges within the brain, independently of fluctuations of their respective values in the bloodstream (Dratman, et al., 1983), suggesting that even small changes within the brain may have major impacts on behavior (Loosen, 1992). Second, T_3 in brain is mostly derived from circulating T_4 through local enzymatic deiodination (5'-D-II diiodinase), rather than through active transport of circulating T_3 into brain tissue (Crantz, et al., 1982). Finally, thyroid hormones in a number of animal studies were shown to reduce the expression of the β -amyloid precursor protein gene (Belandia, et al., 1998).

However, other external factors may influence the level of thyroxine and the association between thyroxine and TSH in serum. In fact, the major part of T_4 (>99%) does not circulate freely but rather binds to thyroxine-binding globulin, thyroxine-binding prealbumin, and albumin, and thus the active form of T_4 is accounted only by 1% of total T_4 . Serum level of thyroxine-binding proteins is greatly determined by other factors in older persons including estrogens, corticosteroid use and liver or renal disease (Surks, et al., 1990). Thus, our findings may be confounded by those external factors, especially in the older age group. Moreover, among healthy participants, the association between thyroxine and TSH was shown to be inverse log-linear. In our study this was the case, with a weak inverse but significant association ($r \sim 0.20$). This weak association suggests that other factors are at play, including the effect of several neurotransmitters on thyroid hormones (somatostatin, cortisol, and cytokines) (Robbins, 1996) and the reduced response of TSH to thyroid releasing hormone with aging among others (Mariotti, et al., 1995).

Similarly, a number of biological mechanisms were suggested to explain leptin's association with cognition. First, leptin may play a role in hippocampal synaptic plasticity that is related to learning and memory and is implicated in leptin induced long-term potentiation (LTP) (Harvey, et al., 2006, Shanley, et al., 2001). Second, it regulates neuron excitability by modulating cognitive ability through AMPK signaling pathway (Harvey, et al., 2006). Third, leptin may act as an anti-apoptotic agent under stress condition and have neuro-protective function (Doherty, et al., 2008, Guo, et al., 2008, Weng, et al., 2007, Zhang, et al., 2007). Fourth, leptin was shown to promote Apolipoprotein-E dependent beta-amyloid re-uptake into the cell, thus reducing its deposition in the extracellular space (Fewlass, et al., 2004). Finally, leptin and insulin act synergistically to reduce the amount of hyperphosphorylation of tau, the main component of the neurofibrillary tangle which constitutes a major hallmark

for AD (Greco, et al., 2008). Importantly, treatment of transgenic mice (AD model) with leptin was shown to trigger improvements in memory tasks (Tezapsidis, et al., 2009).

The antagonistic interaction between thyroxine and TSH may be explained by the fact that mild hypothyroidism, with a prevalence of 5–10% in the general population, is a condition in which T_3 and T_4 are in the normal range while TSH is high. This condition was linked to a number of signs and symptoms including fatigue, poor memory and a slow thinking process, particularly among older women (Canaris, et al., 2000, Evered, et al., 1973, McDermott and Ridgway, 2001). Similarly, the antagonistic interaction between leptin and thyroxine may be due to the function of megalin (also called low-density lipoprotein receptor-related protein-2 [LRP2]), which is an endocytic receptor expressed in a number of epithelial cells including those of the choroid plexus (i.e. blood-brain barrier) and belongs to the low-density lipoprotein (LDL) receptor family. Megalin mediates the transport of both leptin and thyroxine through the blood-brain barrier (Dietrich, et al., 2008, Lisi, et al., 2005), and thus those two hormones may be competing for the same receptor to enter the brain.

Our study has many strengths, including the use of a large nationally representative sample, the inclusion of a wide age range of adults (20–59y and 60–90y) and tests that span a variety of cognitive domains. However, our study is limited because it is cross-sectional so no causal link can be ascertained between the exposures and outcomes. Thus, prospective cohort studies, and ultimately a randomized controlled trial, are needed to better examine the effects of baseline serum leptin, thyroxine and TSH on decline in various domains of cognition and for different age groups. Another limitation is that the type of cognitive tests differed between the two age groups despite spanning a number of domains in both, which did not allow to examine effect modification of the main associations of interest by age per se. In addition, the lack of data on free thyroxine (T_3 or T_4), or thyroxine binding globulin is a major limitation and does not allow for comparison with many of the previous studies which have used those measures instead of total thyroxine level. This has also precluded forming clinically relevant categories that were based on free thyroxine and TSH for hypo and hyperthyroidism as was done previously by others (Gussekloo, et al., 2004). Finally, residual confounding cannot be ruled out even though major potential confounders were adjusted for in our analyses.

In conclusion, our study findings suggest that there are significant associations of serum leptin, thyroxine and TSH with cognitive function among adults, though some of those associations (particularly those of thyroid function) differ markedly between the two age groups of interest. It is important to conduct prospective cohort studies to examine age-related cognitive decline in relation to exposures of interest that would provide further insight into potential interventions that would assess interactive effects of various hormonal replacement regimens.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Figure 1. Study sample selection

Abbreviations: NHANES=National Health and Nutrition Examination Surveys;

TSH=Thyroid Stimulating Hormone.

Figure 2. Dot plot of Log_e transformed leptin, thyroxine and TSH by age group and sex; NHANES III

Abbreviations: ANOVA=Analysis of variance; NHANES=National Health and Nutrition Examination Surveys; T_4 =Thyroxine; TSH=Thyroid Stimulating Hormone.

* $p < 0.0001$ for ANOVA test comparing mean Log_e transformed leptin, T_4 and TSH across age-sex groups. All bonferroni-corrected multiple comparisons were statistically significant ($p < 0.001$), except for $\text{Log}_e(\text{leptin})$ comparing men 20–59yo to men 60–90yo and $\text{Log}_e(\text{TSH})$ comparing 20–59yo men to 60–90yo women.

Graphs by age group and race

Figure 3. Dot plot of Log_e transformed leptin, thyroxine and TSH by age group and race; NHANES III

Abbreviations: ANOVA=Analysis of variance; MA=Mexican-American; NHANES=National Health and Nutrition Examination Surveys; NHB=Non-Hispanic Black; NHW=Non-Hispanic White; OTH=Other ethnicity; T_4 =Thyroxine; TSH=Thyroid Stimulating Hormone.

* $p < 0.0001$ for ANOVA test comparing mean Log_e transformed leptin, T_4 and TSH across age-race groups. All bonferroni-corrected multiple comparisons were statistically significant ($p < 0.05$), except for the following:

$\text{Log}_e(\text{leptin})$: comparing NHW 20–59yo to OTH 20–59yo; comparing NHB 20–59yo to MA 60–90yo; comparing NHW 60–90yo to MA 60–90yo; comparing OTH 60–90yo to NHB 20–59yo, OTH 20–59yo, NHW 60–90yo and NHB 60–90yo; comparing OTH 60–90yo to MA 60–90yo.

$\text{Log}_e(T_4)$: comparing NHW 20–59yo to NHB 20–59yo, NHW 60–90yo and OTH 60–90yo; comparing OTH 20–59yo to 60–90yo(NHB, MA, OTH); comparing NHW 60–90yo to NHB, MA and OTH 60–90yo; comparing NHB 60–90yo to MA and OTH 60–90yo; comparing MA 60–90yo to OTH 60–90yo.

$\text{Log}_e(\text{TSH})$: comparing NHW 20–59yo to MA 60–90yo; comparing OTH 60–90yo to NHW 20–59yo, NHW 60–90yo, MA 20–59yo, and OTH 20–59yo; comparing MA 60–90yo to OTH 60–90yo.

TABLE 1

Study sample characteristics by age group; NHANES III

	20–59y			60–90y		
	N	Mean or %	SE	N	Mean or %	SE
Age						
Female, %	3241	36.7	0.3	6033	72.5	1.6
	3241	51.5	1.1	6033	45.2	7.7
	3241			6033		
Race/ethnicity, %						
NH White		75.0	2.7		82.6	6.0
NH Black		12.0	1.5		13.0	5.9
Mexican-American		5.8	1.2		1.7	0.4
Other ethnicity		7.2	1.6		2.7	7.3
Married, %	3241	60.4	2.1	6033	67.4	5.4
Education, %	3241			6009		
<High School		7.6	1.0		26.7	6.1
High School		48.2	2.0		36.6	6.5
>High School		44.2	2.4		36.7	10.0
Poverty income ratio, %	3241			6033		
0–100%		11.7	1.1		8.4	1.5
>100%–200%		18.3	1.7		26.3	6.1
>200%		70.0	2.1		65.3	6.5
Language of interview	3241			6014		
English		94.8	1.2		98.6	0.3
Spanish		5.2	1.2		1.4	0.3
Cigarette smoking status	3241			6033		
Never		45.7	1.6		35.0	6.0
Former		20.8	1.1		46.3	8.7
Current		33.5	1.7		18.7	5.8
Self-rated health	3241			6033		
Excellent		23.4	1.4		10.2	2.1
Very good		31.6	1.6		33.2	10.4
Good		32.2	1.3		33.5	6.9

	20-59y			60-90y		
	N	Mean or %	SE	N	Mean or %	SE
Fair		11.0	1.1		16.5	2.9
Poor		1.9	0.4		6.3	1.2
1995-Healthy Eating Index, %	3177			5875		
Mean		61.8	0.5		65.7	1.6
Q1: poor quality diet		6.4	0.6		9.6	5.9
Q5: better quality diet		16.1	1.2		28.0	4.9
Physical activity	3183			5921		
More active		22.0	1.4		13.5	2.4
About the same		47.0	1.6		38.8	7.1
Less active		31.0	1.4		47.7	8.5
Co-morbidity index, Mean	3205	1.07	0.03	6025	2.6	0.1
Body mass index, kg.m⁻², Mean	3231	26.2	0.2	6012	26.6	0.1
Leptin level, Log_e-transformed	1293	1.94	0.06	1673	2.25	0.03
T ₁		R: -0.69;1.79	M:1.13		R: -0.22;1.79	M:1.33
T ₂		R:1.81;2.83	M:2.32		R:1.81;2.83	M:2.32
T ₃		R:2.83;4.33	M:3.19		R:2.83;4.55	M:3.20
Thyroxine level, Log_e-transformed	3241	2.67	0.06	6033	2.93	0.19
T ₁		R: -0.92; 2.06	M:1.94		R: -0.92;2.07	M:2.00
T ₂		R: 2.08;2.20	M:2.14		R:2.08;2.20	M:2.19
T ₃		R:2.21;3.40	M:2.32		R:2.21;2.92	M:2.31
Thyroid stimulating hormone, Log_e-transformed	2976	0.32	0.02	5671	0.47	0.13
T ₁		R: -4.60;0.18	M: -0.13		R: -4.60;0.18	M: -0.25
T ₂		R:0.22;0.77	M:0.47		R:0.22;0.77	M:0.47
T ₃		R:0.79;5.50	M:1.03		R:0.79;4.76	M:1.03
Cognitive performance scores						
SRT	2928	233.4	1.5	—	—	—
SDS-L	2895	22.8	0.3	—	—	—
SDS-E	2895	1.4	0.1	—	—	—
SDL-TTC	2807	4.5	0.2	—	—	—

	20–59y			60–90y		
	N	Mean or %	SE	N	Mean or %	SE
SDL-TE	2739	4.6	0.1	—	—	—
WR-CORR		—	—	4674	5.42	0.07
WR-TRIALS		—	—	5754	0.04	0.01
SR-CORR		—	—	5878	3.38	0.54
MATH-INC		—	—	5989	1.51	0.46
ORIENT-INC		—	—	5615	1.88	0.02

Abbreviations: M=Median; MATH-INC=Math test-Incorrect; NH=Non-Hispanic; NHANES=National Health and Nutrition Examination Surveys; OLS=Ordinary Least Square; ORIENT-INC=Orientation to time, incorrect items; SDS-L=Symbol Digits Substitution tests-Latencies; SDS-E=Symbol Digits Substitution tests-Errors; Serial Digits Learning-Trials to criterion; SDL-TE=Serial Digits Learning-Total Errors; SE=Standard error; SR-CORR=Story recall-correct; SRT=Simple reaction time; T=tertile; WR-CORR=Word Recall-Correct; WR-TRIALS=Word Recall-Number of Trials.

TABLE 2

Multiple OLS regression analysis for predictors of serum leptin, thyroxine and thyroid stimulating hormone, by age group; NHANES III

	20–59y				60–90y			
	Log _e (Leptin)		Log _e (Thyroxine)		Log _e (Leptin)		Log _e (Thyroxine)	
	β	SEE	β	SEE	β	SEE	β	SEE
Age	N=1265	N=3141	N=2889	N=1628	N=5856	N=5523		
	0.010	0.003 *	-0.005	0.004	0.009	0.002 *	-0.004	0.003
Female	1.126	0.050 *	-0.047	0.108	-0.067	0.039	0.871	0.045 *
Race/ethnicity (Ref=NH White)								
NH Black	0.009	0.045	0.428	0.125 *	-0.231	0.040 *	0.007	0.060
Mexican-American	0.084	0.071	0.167	0.136	-0.036	0.054	-0.002	0.114
Other ethnicity	0.118	0.117	-0.327	0.129 *	-0.078	0.091	0.209	0.174
Married (Ref=unmarried)	0.055	0.055	0.059	0.094	-0.046	0.034	0.116	0.057 *
Education (Ref High School)								
High School	-0.037	0.093	-0.006	0.207	0.111	0.071	0.091	0.080
>High School	0.052	0.109	0.033	0.180	0.051	0.082	0.017	0.087
Poverty income ratio (Ref=0–100%)								
>100%–200%	-0.035	0.047	-0.290	0.183	-0.044	0.059	-0.060	0.078
>200%	-0.014	0.048	-0.280	0.206	-0.026	0.052	0.000	0.083
Language of interview (Ref=English)								
Spanish	-0.069	0.100	0.066	0.155	0.125	0.088	-0.115	0.167
Cigarette smoking status (Ref=Never)								
Former	0.023	0.057	0.077	0.130	-0.013	0.052	0.085	0.064
Current	-0.076	0.053	0.006	0.110	-0.189	0.051 *	-0.052	0.080
Missing	—	—	-0.429	0.228	-0.716	0.118 *	—	—
Self-rated health (Ref=Excellent)								
Very good	0.089	0.056	0.090	0.122	0.028	0.046	0.152	0.054 *
Good	0.079	0.072	0.140	0.111	-0.065	0.055	0.132	0.072
Fair	0.034	0.064	0.121	0.165	0.071	0.087	0.159	0.070 *
							0.002	0.180
							-0.176	0.073 *
							-0.026	0.065
							-0.167	0.093

	20–59y						60–90y					
	Log _e (Leptin)			Log _e (Thyroxine)			Log _e (Leptin)			Log _e (Thyroxine)		
	β	SE	SEE	β	SE	SEE	β	SE	SEE	β	SE	SEE
Poor	0.143	0.112		0.416	0.440		0.096	0.148		0.586	0.247	
Missing	—	—	—	—	—	—	−0.370	0.094*		1.485	1.747	
1995-Healthy Eating Index (Ref=Q₁)												
Q ₂	0.158	0.123		−0.204	0.202		−0.019	0.124		0.920	0.273*	
Q ₃	0.096	0.123		0.049	0.203		−0.132	0.100		0.544	0.235*	
Q ₄	0.059	0.120		0.034	0.186		−0.108	0.102		0.807	0.244*	
Q ₅ : better quality diet	0.089	0.126		−0.043	0.217		−0.056	0.101		0.920	0.255*	
Missing	0.130	0.200		−0.007	0.278		−0.285	0.136*		0.928	0.366*	
Physical activity (Ref=More active)												
About the same	−0.190	0.060*		−0.111	0.108		0.059	0.075		−0.218	0.194	
Less active	−0.316	0.066*		−0.095	0.120		−0.004	0.080		−0.446	0.252	
Body mass index, kg.m^{−2}	0.099	0.005*		−0.006	0.008		0.090	0.006*		−0.034	0.015*	
Co-morbidity index	−0.035	0.015*		0.110	0.055		0.017	0.013		−0.019	0.040	

* P<0.05 for null hypothesis that β=0.

Abbreviations: NH=Non-Hispanic; NHANES=National Health and Nutrition Examination Surveys; OLS=Ordinary Least Square; SE=Standard error; Q=Quintile.

TABLE 3

Multiple OLS and zero-inflated poisson regression analysis for predictors (thyroxine, TSH and leptin) of cognitive performance, by age group; NHANES III

20-59y											
SRT			SDS-L			SDS-E			SDL-TTC		
β	SEE	β	SEE	β	SEE	β	SEE	β	SEE	β	SEE
Model 1: Leptin[†]											
N=1156		N=1144		N=1144		N=1144		N=1108		N=1131	
T ₂ vs. T ₁	5.132	4.703	0.023	0.630	-0.768	0.255*	0.137	0.291	0.117	0.476	
T ₃ vs. T ₁	-8.752	7.693	-0.754	0.686	-0.198	0.424	-0.072	0.431	-0.008	0.720	
Model 2: Thyroxine[†]											
N=2617		N=2593		N=2593		N=2593		N=2474		N=2526	
T ₂ vs. T ₁	-1.870	2.778	-0.102	0.253	0.192	0.150	0.285	0.133*	0.334	0.241	
T ₃ vs. T ₁	-1.476	2.407	-0.456	0.299	0.190	0.103	-0.003	0.147	-0.166	0.263	
Model 3: TSH[†]											
N=2639		N=2611		N=2611		N=2611		N=2483		N=2545	
T ₂ vs. T ₁	1.011	2.681	0.303	0.290	0.236	0.131	-0.048	0.138	0.080	0.273	
T ₃ vs. T ₁	7.052	3.299*	0.803	0.310*	0.134	0.181	-0.007	0.180	0.132	0.317	
60-90y											
WR-CORR			WR-TRIALS			SR-CORR			MATH-INC		
β	SEE	β	SEE	β	SEE	β	SEE	β	SEE	β	SEE
Model 1: Leptin[†]											
N=1372		N=1362		N=1416		N=1468		N=1389			
T ₂ vs. T ₁	-0.022	0.016	0.722	0.467	-0.004	0.039	0.127	0.136	-0.001	0.027	
T ₃ vs. T ₁	-0.026	0.021	0.902	0.943	-0.014	0.051	-0.060	0.147	-0.029	0.039	
Model 2: Thyroxine[†]											
N=4077		N=5186		N=5270		N=5317		N=5080			
T ₂ vs. T ₁	0.007	0.006	0.113	0.315	0.113	0.070	-0.220	0.101*	-0.004	0.013	
T ₃ vs. T ₁	-0.011	0.010	0.040	0.346	0.142	0.070*	-0.247	0.110*	0.001	0.014	
Model 3: TSH[†]											
N=4236		N=5328		N=5437		N=5499		N=5232			
T ₂ vs. T ₁	0.005	0.009	0.220	0.438	0.219	0.119	-0.312	0.129*	-0.030	0.014*	

20–59y									
SRT		SDS-L		SDS-E		SDL-TTC		SDL-TE	
	β	SEE	β	SEE	β	SEE	β	SEE	β
T ₃ vs. T ₁	0.015	0.006*	−0.031	0.405	0.291	0.120*	−0.265	0.091*	−0.019
									0.009

* P<0.05 for null hypothesis that $\beta=0$;

$\dagger$ P<0.10 for null hypothesis that sex $\times$ exposure (leptin, thyroxine, TSH) interaction term $\gamma=0$ in model with sex among main effects;

$\S$ P<0.10 for null hypothesis that race $\times$ exposure (leptin, thyroxine, TSH) interaction term $\gamma=0$ in model with race among main effects.

Abbreviations: MATH-INC=Math test-Incorrect; NH=Non-Hispanic; NHANES=National Health and Nutrition Examination Surveys; OLS=Ordinary Least Square; ORIENT-INC=Orientation to time, incorrect items; SDS-L=Symbol Digits Substitution tests-Latencies; SDS-E=Symbol Digits Substitution tests-Errors; Serial Digits Learning-Trials to criterion; SDL-TE=Serial Digits Learning-Total Errors; SE=Standard error; SR-CORR=Story recall-correct; SRT=Simple reaction time; WR-CORR=Word Recall-Correct; WR-TRIALS=Word Recall-Number of Trials.

I All three predictors (leptin, thyroxine and TSH) were first Log_e transformed and then converted to tertiles. Models were adjusted for age (continuous), sex, race, marital status (married vs. not); education (<High School; High School; >High School), poverty income ratio (0–100%; >100–200%; >200%), language of interview (English vs. Spanish), cigarette smoking status (never, former, current), self-rated health (excellent, very good, good, fair, poor), 1995-healthy eating index (quintiles), physical activity (more active, about the same, less active), body mass index and a co-morbidity index. Models for the 20–59y age group were multiple OLS regression models, while those for the 60–90y age group were poisson or zero-inflated poisson regression models.

TABLE 4

Multiple OLS and zero-inflated poisson regression analysis for predictors (thyroxine, TSH and leptin) of cognitive performance: two-way interactions between exposure variables (tertiles), by age group; NHANES III

	20-59y											
	SRT			SDS-L			SDS-E			SDL-TTC		
	β	SEE	β	β	SEE	β	β	SEE	β	β	SEE	β
<i>Model 1: I</i>	N=2591		N=2567			N=2567			N=2449			N=2501
Thyroxine												
T ₂ vs. T ₁	0.440	2.881	0.296	0.299	0.097	0.148	0.325	0.129*	0.566	0.229*		
T ₃ vs. T ₁	2.577	3.044	0.303	0.440	0.072	0.268	0.089	0.194	0.338	0.319		
TSH												
T ₂ vs. T ₁	2.905	2.919	0.721	0.317*	0.161	0.158	0.008	0.145	0.358	0.296		
T ₃ vs. T ₁	10.244	4.300*	1.626	0.511*	-0.036	0.314	0.087	0.237	0.641	0.399		
Thyroxine×TSH	-2.266	1.738	-0.430	0.213*	0.087	0.135	-0.047	0.093	-0.289	0.1141*		
<i>Model 2: I</i>	N=1154		N=1142			N=1142			N=1107			N=1129
Leptin												
T ₂ vs. T ₁	7.650	6.315	-0.629	0.736	-0.790	0.289*	0.051	0.324	0.036	0.572		
T ₃ vs. T ₁	-6.759	7.630	-1.177	0.782	-0.221	0.380	-0.127	0.466	-0.010	0.840		
Thyroxine												
T ₂ vs. T ₁	-3.897	5.931	-0.804	0.642	0.180	0.357	-0.031	0.217	0.452	0.401		
T ₃ vs. T ₁	0.138	4.885	-1.359	0.476*	-0.023	0.197	-0.229	0.263	-0.728	0.543		
Thyroxine×Leptin	-0.510	0.920	0.113	0.086	0.005	0.043	0.015	0.037	0.008	0.069		
<i>Model 3: I</i>	N=1143		N=1131			N=1131			N=1098			N=1118
Leptin												
T ₂ vs. T ₁	2.808	5.219	-0.656	0.902	-0.579	0.347	-0.493	0.356	-0.996	0.553		
T ₃ vs. T ₁	-10.981	6.881	-1.127	0.793	-0.035	0.458	-0.496	0.496	-0.718	0.819		
TSH												
T ₂ vs. T ₁	1.318	4.644	-0.770	0.797	0.134	0.310	-0.529	0.249*	0.894	0.431*		

20-59y											
	SRT		SDS-L		SDS-E		SDL-TTC		SDL-TE		
	β	SEE	β	SEE	β	SEE	β	SEE	β	SEE	
T ₃ vs. T ₁	9.475	6.115	-0.242	0.824	0.043	0.368	-0.560	0.239*	0.504	0.494	
TSH×Leptin	0.332	0.921	0.156	0.124	-0.007	0.052	0.143	0.043*	0.248	0.073*	
60-90y											
	WR-CORR		WR-TRIALS		SR-CORR		MATH-INC		ORIENT-INC		
	β	SEE	β	SEE	β	SEE	β	SEE	β	SEE	
Model 1: /	N=4005		N=5115		N=5196		N=5244		N=5020		
Thyroxine											
T ₂ vs. T ₁	-0.011	0.011	0.062	0.542	0.422	0.196*	-0.536	0.115*	-0.016	0.016	
T ₃ vs. T ₁	-0.007	0.019	-0.121	0.695	0.643	0.279*	-0.842	0.152*	-0.023	0.023	
TSH											
T ₂ vs. T ₁	0.007	0.009	0.243	0.471	0.389	0.195~	-0.459	0.140*	-0.036	0.017*	
T ₃ vs. T ₁	0.020	0.012	-0.147	0.559	0.655	0.279*	-0.591	0.126*	-0.035	0.016*	
Thyroxine×TSH	-0.001	0.007	0.040	0.232	-0.198	0.086*	+0.237	0.052*	+0.011	0.010	
Model 2: /	N=1356		N=1346		N=1400		N=1450		N=1374		
Leptin											
T ₂ vs. T ₁	0.010	0.023	-0.064	1.005	0.002	0.045	0.067	0.162	-0.008	0.032	
T ₃ vs. T ₁	0.004	0.025	-0.316	1.064	0.005	0.068	-0.078	0.169	-0.020	0.038	
Thyroxine											
T ₂ vs. T ₁	0.041	0.023	-0.062	1.437	-0.011	0.052	-0.083	0.221	-0.010	0.036	
T ₃ vs. T ₁	0.028	0.018	0.297	0.774	-0.042	0.071	-0.095	0.143	-0.022	0.029	
Thyroxine×Leptin	-0.008	0.003*	0.148	0.170	0.002	0.011	0.019	0.026	0.002	0.006	
Model 3: /	N=1348		N=1338		N=1390		N=1442		N=1364		
Leptin											
T ₂ vs. T ₁	-0.009	0.034	6.213	2.886*	-0.001	0.059	0.458	0.168*	-0.033	0.038	

	20-59y									
	SRT		SDS-L		SDS-E		SDL-TTC		SDL-TE	
	β	SEE	β	SEE	β	SEE	β	SEE	β	SEE
T ₃ vs. T ₁	-0.007	0.031	5.258	2.008*	-0.004	0.063	0.066	0.665	-0.043	0.037
TSH										
T ₂ vs. T ₁	0.017	0.029	6.792	2.461*	0.002	0.059	0.324	0.185~	-0.037	0.045
T ₃ vs. T ₁	0.021	0.024	4.607	1.689*	0.047	0.045	0.261	0.170	-0.028	0.037
TSH×Leptin	-0.003	0.004	-0.679	0.348~	-0.001	0.009	-0.057	0.038	0.003	0.006

~P<0.10

* P<0.05 for null hypothesis that $\beta=0$.

Abbreviations. MATH-INC=Math test-Incorrect; NH=Non-Hispanic; NHANES=National Health and Nutrition Examination Surveys; OLS=Ordinary Least Square; ORIENT-INC=Orientation to time, incorrect items; SDS-L=Symbol Digits Substitution tests-Latencies; SDS-E=Symbol Digits Substitution tests-Errors; Serial Digits Learning-Trials to criterion; SDL-TE=Serial Digits Learning-Total Errors; SE=Standard error; SR-CORR=Story recall-correct; SRT=Simple reaction time; WR-CORR=Word Recall-Correct; WR-TRIALS=Word Recall-Number of Trials.

J All three predictors (leptin, thyroxine and TSH) were first Loge transformed and then converted to tertiles. Models were adjusted for age (continuous), sex, race, marital status (married vs. not); education (<High School; High School; >High School), poverty income ratio (0-100%; >100-200%; >200%), language of interview (English vs. Spanish), cigarette smoking status (never, former, current), self-rated health (excellent, very good, good, fair, poor), 1995-healthy eating index (quintiles), physical activity (more active, about the same, less active), body mass index and a co-morbidity index. Models for the 20-59y age group were multiple OLS regression models, while those for the 60-90y age group were poisson or zero-inflated poisson regression models.