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ORIGINAL ARTICLE

TSH concentration within the normal range is associated with cognitive function and ADHD symptoms in healthy preschoolers

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Summary

Objective Thyroid hormone concentrations outside the normal range affect brain development, but their specific influence on behaviour and mental abilities within normal values is unknown. The objective of this study was to investigate whether thyroid hormone concentrations are related to neurodevelopment and ADHD (attention deficit and hyperactivity disorder) symptoms in healthy preschoolers.

Design subjects and measurements Children from two general population birth cohorts in Menorca ($n = 289$) and Ribera d'Ebre ($n = 53$), Spain, were assessed in a cross-sectional study at the age of 4. Thyroid hormones (free T4 and T3) and TSH concentrations were measured and mental and motor development was assessed using McCarthy's scales for neuropsychological outcomes and ADHD-DSM-IV for attention deficit and hyperactivity/impulsivity symptoms.

Results Children with TSH concentrations in the upper quartile of the normal range performed lower on McCarthy's scales and were at higher risk for attention deficit and hyperactivity/impulsivity symptoms. In the Menorca cohort, a decrease of 5·8 ($P < 0·05$) and 6·9 ($P < 0·01$) points was observed in memory and quantitative skills, respectively. In contrast, high T4 concentrations were associated with decreased risk of having 1–5 attention deficit symptoms (odds ratio: 0·25; $P < 0·01$); these findings were observed in both cohorts despite differences in mean TSH concentrations. No associations were observed with T3.

Conclusions Despite being within the normal range, high TSH concentrations are associated with a lower cognitive function and high TSH and low free T4 with ADHD symptoms in healthy preschoolers. Statistically significant differences were observed in the highest quartiles of TSH, suggesting a need for re-evaluation of the upper limit of the normal TSH range.

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Introduction

Thyroid hormones (TH) are essential for normal development of the human foetal brain.[1] They regulate the processes of dendritic and axonal growth, synaptogenesis, neuronal migration, and myelination.[2–4] Congenital hypothyroidism is a lack of TH in the newborn, due to an absence or abnormal development of the thyroid gland, destruction of the thyroid gland, failure of stimulation of the thyroid by the pituitary gland, or defective or abnormal formation of thyroid hormones, which can result in severe mental and physical retardation, known as cretinism in humans.[4,5] It has been described in several studies that maternal iodine deficiency or mild hypothyroidism during pregnancy is associated with lower IQ levels in the child,[6,7] because the foetus is dependent on maternal thyroxine during the first two trimesters.[1,8] Studies among children with congenital hypothyroidism have also shown a relationship between concentrations of TH at birth and neurodevelopment.[9–18] Nevertheless, early treatment of congenital hypothyroidism partially prevents intellectual impairment.[17,18] Even if treated, children with congenital hypothyroidism frequently demonstrate impaired neuromotor,[9] visuospatial,[11,13] memory,[12,13] verbal,[9,12,13] and attention skills.[12,13]

Thyroid hormones are also important for the behaviour and cognitive function of the child and the adult brain;[4,19] however, little is known about their mechanisms of action. Some mechanisms have been proposed,[20–26] suggesting a predominant effect of TH in the hippocampus and prefrontal cortex, involving memory and attention functions. In general, patients with hypothyroidism show impaired cognition and generalized neural dysfunction while hyperthyroid patients show irritability and anxiety.[27,28] More detailed studies using psycho-behavioural testing in hypothyroid patients have revealed deficits in memory and learning, attention, motor speed, visuoperceptual and construction skills.[29] Nonetheless, there are no studies that evaluate the role of thyroid function in

variations of memory, attention, or other skills, when TH levels are within the normal range for children or adults.

The aim of the present study was to assess the association of TH and thyroid stimulating hormone (TSH) concentrations with cognitive development and attention deficit and hyperactivity/impulsivity symptoms, in healthy children from the general population at 4 years of age.

Methods

Subjects

This study is based on data from two birth-cohorts from the general population: (a) from the Spanish Balearic Island of Menorca, a popular tourist destination located in the northwest Mediterranean sea;[30] and (b) from Ribera d'Ebre, a rural area with an electrochemical plant situated in the northeastern, Catalonia region of Spain.[31] Children from the Menorca cohort were participants in the Asthma Multicentre Infant Cohort Study (AMICS), a European study assessing factors causing asthma in children.[30] The cohort from Ribera d'Ebre was originally established to assess the effect of organochlorines in child development.[31,32] The cohorts from Menorca (482 children born between 1997 and 1998 and Ribera d'Ebre (102 children born between 1997 and 1999) were followed to the age of 4 years for psychometrical and thyroid function assessment during the period 2001–2003. Blood and concentrations of TH and TSH measurements were taken in a total of 342 children (289 and 53, respectively) and were included in the final data analyses. The most common reasons for noninclusion were refusal by parents to give permission for blood being taken or an insufficient quantity of serum to enable measurement of TH and TSH. Children from Ribera d'Ebre cohort included in our final study population ($n = 53$) differed from those not included ($n = 49$) in the McCarthy quantitative scale [means (SD) were 109 (18·6) and 101 (12·1), respectively], and in the proportion of mothers who had smoked at delivery (21 *vs.* 41%, respectively). However, they did not differ in any of the other developmental or maternal variables shown in Table 1 ($P > 0{\cdot}1$). Children included from the Menorca cohort ($n = 289$) did not differ from those not included ($n = 193$) in any of the variables shown in Table 1. The present study cross-sectionally assesses children at the age of 4.

Table 1. Characteristics of the study population

	Menorca ($n = 289$)	Ribera d'Ebre ($n = 53$)
Child variables		
Gestational age (weeks)	39·3	39·6
Sex (%males)	50	45
Breastfeeding (%)	85	80
McCarthy (mean score)		
General cognitive	99	107*
Verbal	99	104*
Perceptual-performance	99	107*
Quantitative	100	109*
Memory	99	105*
Motor	100	104*
ADHD (% with number of attention deficit or hyperactivity symptoms ≥ 6)	15	14
Attention deficit symptoms (% ≥ 6)	11	10
Hyperactivity/impulsivity symptoms (% ≥ 6)	6	6
Maternal variables		
Mean age at delivery (years)	30	30
Social class (%)		
Professional, manager, technician	14	21
Skilled manual and nonmanual	51	11
Partial skilled and unskilled	16	32
Unemployed	19	36*
Years of school completed (% less than primary)	7	46*
Smoking at delivery (%)	21	21
Smoking 4 years after delivery (%)	31	32

*P-value < 0·05.

Neurodevelopmental assessment

The children's mental and psychomotor development was assessed at the age of 4 using the McCarthy scales of children's abilities (MCSA).[33] The MCSA consists of 18 items derived from six different scales (general cognitive, verbal, perceptual-performance, quantitative, memory, and motor). Three neuropsychologists (two for the Menorca cohort and one for the Ribera d'Ebre cohort) were trained to administer and interpret the MCSA. To limit interobserver variability, we applied a strict protocol, including triple-blinded interobserver training comparisons and three triple-blinded interobserver quality control trials undertaken during the fieldwork. The interobserver variability was lower than 5%. Following neuropsychological testing, an attention and hyperactivity questionnaire (ADHD-DSM-IV)[34] was completed by the children's teachers (preschool starts at 3 years of age and the teacher spends approximately 5 h every day with the children). Attention deficit hyperactivity disorder (ADHD) criteria of the *Diagnostic and Statistical Manual of Mental Disorders* (DSM-IV)[34] is a well-known questionnaire composed of 18 items that evaluate attention deficits (1–9), hyperactivity (10–15) and impulsiveness (16–18) in children; each item has only two response options: 'yes' or 'no'. The DSM-IV criteria for the diagnosis of ADHD, defined by presenting six or more attention deficit symptoms, or six or more hyperactivity/impulsivity symptoms, were taken into account.

Thyroid function assessment

Thyroid function of children from the Menorca and Ribera d'Ebre cohorts was assessed at the age of 4 years by measuring the concentrations of TSH, T3 and free T4 in serum samples in the reference laboratory of Catalonia by chemiluminescence assay (Architect system®, Abbott Diagnostics, Madrid, Spain) during 2004. Interassay coefficients of variation (CV) for TSH, free T4 and T3 measurements were under 5·2, 7·8 and 5·3%, respectively, and intra-assay coefficients were 3·3, 4·2 and 3%, respectively. The reference criteria proposed were 0·35–5 mU/l for TSH, 85–250 ng/dl for T3 and 0·7–1·7 ng/dl for free T4. Samples from both cohorts were stored at –20 °C prior to the analyses.

Table 2. Thyroid hormone concentrations in subjects from Menorca and Ribera d'Ebre cohorts at 4 years of age ($n = 342$)

	Min	PC5	PC25	PC50	PC75	PC95	Max	Mean (SD)
Menorca cohort ($n = 289$)								
Free T4 (ng/dl)	0·4	0·85	0·96	1·04	1·14	1·25	1·44	1·04 (1·13)
T3 (ng/dl)	25	94	138	152	165	183	226	151 (22)
TSH (mU/l)	0·45	0·75	1·24	1·68	2·20	3·19	5·01	1·78 (0·76)
Ribera d'Ebre cohort ($n = 53$)								
Free T4 (ng/dl)	0·95	1·02	1·12	1·19	1·27	1·41	1·5	1·20 (0·11)
T3 (ng/dl)	98	130	142	155	183	208	232	162 (27)
TSH (mU/l)	0·45	0·86	1·61	2·64	3·33	5·4	6·04	2·69 (1·3)

Other variables

Information on both the children's and parental education, socioeconomic background (using The UK Registrar General's 1990 classification according to parental occupation by ISCO88 code), demographic factors, marital status, maternal disease and obstetric history, parity, duration of breastfeeding, gender, alcohol consumption during pregnancy, children's exposure to cigarettes, and dietary fish intake was obtained by questionnaire. Gestational age and anthropometric measures at birth were collected from clinical records and anthropometric measures at 4 years were collected ad hoc.

Statistical analyses

The continuous neurodevelopmental outcomes were standardized to a mean of 100 with a standard deviation (SD) of 15. The association between thyroid hormones and TSH and McCarthy's scales was tested using linear regression. Adjusted general additive models (GAM) were used to evaluate the linearity of the relationship between T4 and TSH concentrations and neurodevelopmental outcomes, using nonparametric depiction of the predictor on the outcome over the range of the predictor when the effects of the other variables had been taken into account. In subsequent models, TSH concentration was categorized into quartiles of the distribution of TSH in the entire study population. ADHD scales where categorized into three groups in order to differentiate those children with no symptoms from those with one or more symptoms but with no diagnosis. The third group comprised the children with a disorder diagnosed using the DSM-IV criteria (i.e. > 5 symptoms of ADHD). The associations between quartiles of TSH and free T4, and the different groups of the ADHD scales were evaluated using a multinomial logistic regression model; the group of children with no symptoms was used as the control group. All models were adjusted for maternal social class and level of education during pregnancy, children's gender, age and school term of test administration, and evaluator psychologist. The following were not included in the models because their coefficients were not statistically significant [(P-value > 0·20) in the bivariate analyses]: gestational age, children's cranial perimeter, height and weight at birth, type of delivery, children's weight and height at age 4, mother's passive exposure to tobacco during pregnancy, mother's age at delivery, and father's smoking habits when the child was 4 years of age. In addition, a set of variables potentially related to the outcome, including home family size, mother's smoking habits during and after pregnancy, and breastfeeding was included in the final model if $P < 0·20$ and if the coefficient was modified by more than 10% in the multivariate model.

Results

Table 1 shows the characteristics of the study population by cohort. Children from Ribera d'Ebre tended to perform better on McCarthy's tests. Mothers from Ribera d'Ebre had lower educational levels and a higher unemployment rate.

Table 2 presents the thyroid hormones and TSH concentrations of the study population. Descriptive statistics are reported as percentiles (PC). Children from Menorca had lower concentrations of TSH, free T4, and T3 ($P < 0·05$). None of the children presented any diagnosed thyroid disorder and most of the thyroid hormones and TSH concentrations were within the normal range. However, two children from Menorca each had a free T4 concentration below 0·7 ng/dl, one child from Menorca had T3 concentrations below 85 ng/dl, and five children from Ribera d'Ebre had a TSH concentration slightly above the reference range (5 mU/l) but normal free T4 and T3 concentrations. These children were excluded from subsequent analyses.

The adjusted coefficients for the relationship between TSH concentration, as a continuous trait, and McCarthy's scores are shown in Table 3. TSH concentration was negatively associated with most of the McCarthy scales in the two cohorts, although stronger associations were observed in Ribera d'Ebre. In both cohorts, decreases on the memory and quantitative scales were statistically significant. For instance, decreases of 2·6 ($P < 0·05$) and 6·7 ($P < 0·01$) points in the memory scale were observed in Menorca and Ribera d'Ebre, respectively, for each increase of 1 mU/l of TSH. No statistically significant associations were observed between free T4 or T3 concentrations and McCarthy scales; however, free T4 was positively related to most of McCarthy's scales (data not shown).

Figure 1 depicts the dose–response relationship between TSH and McCarthy's general cognitive and quantitative scores for each cohort. TSH concentration shows a negative relationship in both cohorts, with a stronger decline above concentrations around 2 mU/l. Similarly, there was a consistent linear relationship between the highest TSH concentrations and perceptual-performance (Menorca

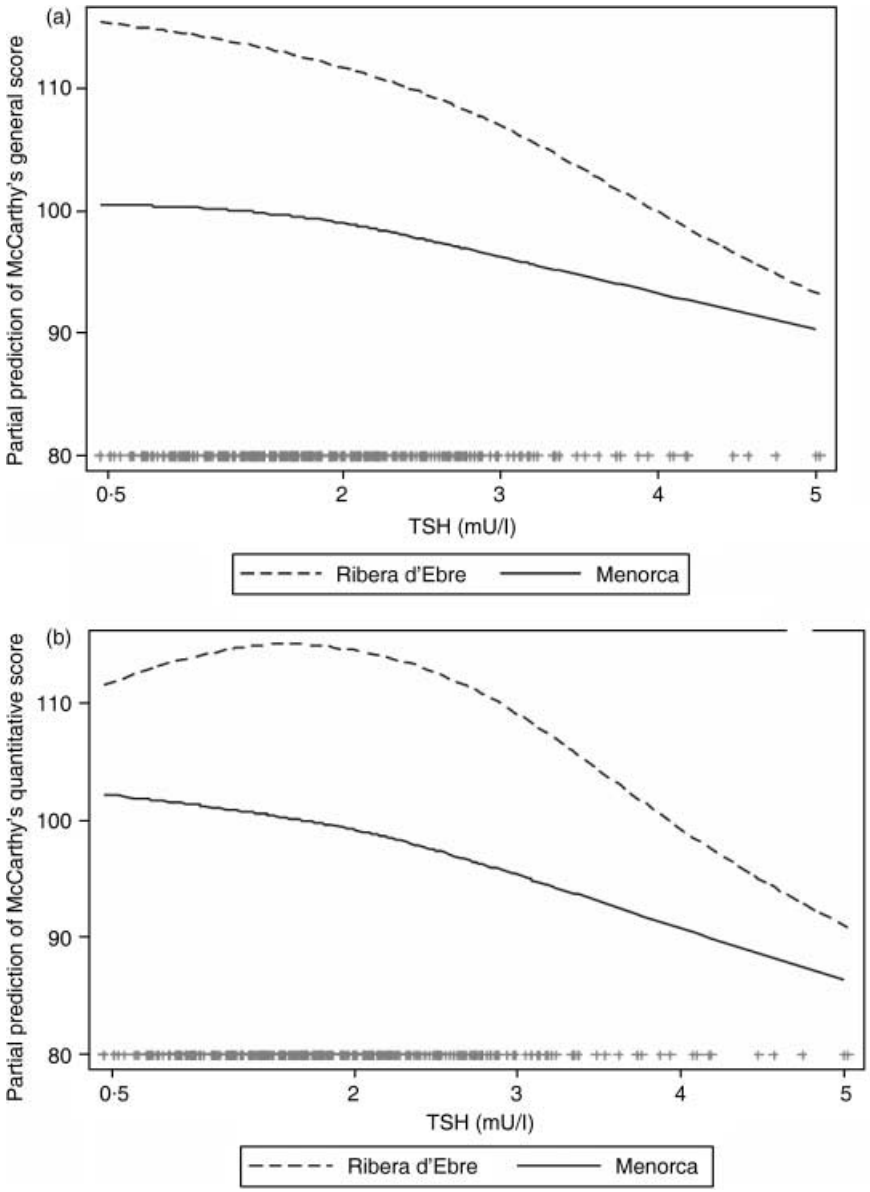

Fig. 1 Adjusted† association between TSH and (a) McCarthy's general cognitive and (b) quantitative scores according to the cohort. (GAM models). †The relation is adjusted for mother's social class and level of education during pregnancy, child's gender, age and school term of test administration, evaluator psychologist, number of brothers/sisters, duration of breastfeeding, and mother's smoking habits.

Table 3. Adjusted† association of TSH levels and neurodevelopmental outcomes at 4th year of age. All numbers are coefficients (standard error)

	Menorca (n = 286)	Ribera d'Ebre (n = 48)
McCarthy scale		
General cognitive	–2·1 (1·1)	–5·4 (2·2)*
Verbal	–1·1 (1·1)	–6·4 (1·9)**
Perceptual-performance	–1·9 (1·1)	0·36 (2·6)
Quantitative	–2·9 (1·1)**	–5·9 (2·6)*
Memory	–2·6 (1·1)*	–6·7 (2·1)**
Motor	–1·9 (1·0)	3·3 (2·3)

*P-value < 0·05; **P-value < 0·01.
†Models have been adjusted for mother's social class and level of education during pregnancy, child's gender, age and school term of test administration, evaluator psychologist, and the following covariates depending on the model: mother's smoking habit (during pregnancy or at child aged 4 years), number of brothers/sisters and duration of breastfeeding.

cohort), verbal (Ribere d'Ebre cohort) and memory scales (both cohorts) (data not shown). No relationship was observed between free T4 and T3 concentration and McCarthy's outcomes (data not shown).

Table 4 shows the adjusted associations with TSH treated as a categorical variable defined by the quartiles. Children from the fourth quartile performed significantly worse in some of the McCarthy outcomes, with the quantitative and memory scales being the most notably impaired (6·9 and 5·8 points, respectively) in the Menorca cohort, and the verbal and memory scales in the Ribera d'Ebre cohort (20·4 and 19·1 points, respectively). However, there were no differences between the first three quartiles.

The association between quartiles of TSH and freeT4, and the different groups of the ADHD scales in Menorca is shown in Table 5. Only data from the Menorca cohort are shown, because there were few children from Ribera d'Ebre to categorize the dependent and the independent variables. Children from the fourth quartile of TSH,

Table 4. Adjusted† association between TSH in quartiles and neurodevelopmental outcomes in Menorca ($n = 286$) and Ribera d'Ebre cohort ($n = 48$)

	TSH quartiles‡ (mU/l)			
	Reference score	Coefficients (SE)		
	0·45–1·28	1·29–1·70	1·71–2·42	2·43–5·01§
McCarthy scales (Menorca)	$n = 78$	$n = 75$	$n = 76$	$n = 57$
General cognitive	106·5 (3·5)	–1·7 (2·2)	–1·1 (2·3)	–5·4 (2·4)*
Perceptual-performance	104·7 (3·2)	–2·3 (2·2)	–1·9 (2·2)	–5·3 (2·4)*
Quantitative	103·1 (3·4)	–2·1 (2·3)	–1·7 (2·3)	–6·9 (2·4)**
Memory	102·7 (3·6)	–2·3 (2·4)	–1·6 (2·4)	–5·8 (2·6)*
Verbal	107·8 (3·8)	–0·6 (2·4)	–0·3 (2·4)	–2·7 (2·6)
Motor	103·0 (3·0)	–1·3 (2·1)	–4·1 (2·1)	–4·2 (2·3)
McCarthy scales (Ribera d'Ebre)	$n = 6$	$n = 10$	$n = 9$	$n = 23$
General cognitive	120·5 (10·8)	–7·7 (10·9)	–4·6 (11·3)	–12·7 (10·0)
Perceptual-performance	106·1 (9·7)	–4·8 (10·1)	10·0 (11·4)	6·2 (10·0)
Quantitative	103·3 (11·3)	5·8 (11·5)	2·0 (12·6)	–5·6 (11·3)
Memory	120·8 (9·4)	–5·9 (9·0)	–9·8 (10·6)	–19·1 (9·2)*
Verbal	124·1 (9·5)	–12·4 (8·8)	–12·3 (9·9)	–20·4 (8·8)*
Motor	93·5 (8·8)	–2·8 (9·2)	5·8 (10·4)	11·0 (9·1)

*P-value < 0·05; **P-value < 0·01.
†Models have been adjusted for mother's social class and level of education during pregnancy, child's gender, age and school term of test administration, evaluator psychologist and the following covariates depending on the model: mother's smoking habit (during pregnancy or when child aged 4 years), number of brothers/sisters and duration of breastfeeding.
‡Quartiles of pooled levels of TSH concentrations of the two cohorts.
§Five children with TSH > 5·01 mU/l were excluded.

compared to those from the first quartile, had a high risk of having more than six attention deficit symptoms and 1–5 hyperactivity/impulsivity symptoms (odds ratio = 4·9; $P < 0·05$ and odds ratio = 3·2; $P < 0·05$, respectively). In contrast, children with the highest free T4 concentrations had less risk of having symptoms than children from the first quartile did.

Discussion

We observed that high concentrations of TSH were negatively associated with memory and verbal and quantitative skills, as well as positively associated with attention deficit and hyperactivity/impulsivity symptoms. In addition, children with high free T4 concentration had a decreased risk of having poor attention. The effects of TSH and TH on cognitive function were observed in both cohorts despite differences in the distributions of hormone concentrations measured in the two populations. Because all samples were analysed by a single laboratory, the between-cohort differences in TSH and TH are not explainable by laboratory analysis methods or practices. However, they may be a result of differences in dietary iodine intake between the two populations or due to other variables not assessed in this study such as environmental pollution or genetic background. Both communities reported diets rich in fish (the main source of iodine), although urinary iodine excretion is unknown. Indeed, one of the communities has a history of environmental contamination with organochlorine compounds that have been epidemiologically associated with elevated incidence of thyroid cancer[35] and lower levels of total T4.[36] Moreover, children from Ribera d'Ebre presented higher scores on McCarthy's scales, which might be explained by socioeconomic and cultural factors, genetic differences, or by the differences between the psychologists who administered and interpreted the MCSA in each cohort, even though they followed a cross-validation quality control procedure. Nevertheless, impaired mental development in children with higher TSH concentrations was observed in both communities.

The relationship between thyroid hormones or TSH and cognitive function has been studied in children with congenital hypothyroidism,[9–18] children from mothers with low thyroid hormone concentrations during pregnancy,[6,7,37] children who lived in iodine-deficient areas,[38] and in populations with resistance to thyroid hormone syndrome (RTH).[39] This relationship has also been studied in populations with some neurological alteration such as dementia in adults,[40] ADHD in children,[41,42] and in populations with depression.[43] However, no studies have reported the association between TH concentrations and neurodevelopmental outcomes in children from the general population with nonpathological values of thyroid hormone concentrations and without neurological alterations.

Despite differences in some characteristics of the base populations, the results of our study are consistent with the results of other clinical studies. A study among children at a clinic specializing in learning and behaviour problems found an association between low concentration of free T4 and ADHD.[41] Many longitudinal studies of congenital hypothyroidism have indicated a correlation between

Table 5. Adjusted§ association (odds ratio and 95%CI) of TSH and T4 quartiles and symptoms of attention deficit and hyperactivity/impulsivity disorder (ADHD) in Menorca ($n = 286$)

	TSH quartiles‡			Free T4 quartiles‡		
	2nd (TSH = 1·25–1·68)	3rd (TSH = 1·69–2·20)	4th (TSH = 2·21–5·01)	2nd (T4 = 0·97–1·04)	3rd (T4 = 1·05–1·14)	4th (T4 = 1·15–1·44)
ADHD†						
1–5 symptoms of attention deficit and hyperactivity/impulsivity	0·68 (0·27–1·69)	1·54 (0·65–3·63)	1·62 (0·62–4·2)	0·88 (0·37–2·09)	1·03 (0·43–2·44)	0·34 (0·13–0·89)*
More than 6 symptoms of attention deficit or hyperactivity/impulsivity	1·15 (0·34–8·84)	1·00 (0·27–3·70)	2·80 (0·80–9·88)	1·59 (0·54–4·67)	0·66 (0·9–2·23)	0·51 (0·15–1·76)
Attention deficit†						
1–5 attention deficit symptoms	1·04 (0·42–2·62)	1·72 (0·72–4·11)	1·98 (0·76–5·18)	1·58 (0·25–1·34)	0·75 (0·33–1·72)	0·25 (0·09–0·66)**
More than six symptoms of attention deficit	1·69 (0·38–7·47)	0·50 (0·07–3·30)	4·94 (1·05–23·16)*	1·60 (0·46–5·55)	0·43 (0·10–1·91)	0·42 (0·10–1·80)
Hyperactivity/impulsivity†						
1–5 symptoms of hyperactivity/impulsivity	1·68 (0·65–4·37)	2·42 (0·45–6·20)	3·17 (1·16–8·66)*	0·74 (0·31–1·76)	0·96 (0·40–2·33)	0·56 (0·22–1·45)
More than six symptoms of hyperactivity/impulsivity	3·25 (0·53–29·86)	2·17 (0·34–13·80)	3·30 (0·50–21·63)	1·58 (0·44–5·69)	0·82 (0·18–3·77)	0·82 (0·17–3·92)

†The reference category is the group of children with no symptoms. ‡The first quartile is the reference category.
*P-value < 0·05; **P-value < 0·01.
§Models have been adjusted for mother's social class and level of education during pregnancy, child's gender, age and school term of test administration, evaluator psychologist, and the following covariates depending on the model: mother's smoking habit (during pregnancy or at child aged 4 years), number of brothers/sisters and duration of breastfeeding.

severity of hypothyroidism and neurodevelopmental outcomes.[8–17] Rovet observed that verbal and memory competencies were particularly affected in children with congenital hypothyroidism.[12,13] Another study assessed the neuropsychological development in 7-year-old children born to mothers with the highest TSH concentrations during pregnancy in a large population, but who were untreated; these children averaged 7 points lower in the full-scale IQ scores.[6] The impact of thyroid function on cognitive ability has also been observed in old age. Wahlin *et al.*[44,45] reported that TSH within normal ranges, but not free T4, predicted cognitive performance in cross-sectional and longitudinal assessments of individuals aged 75–96 years.

Most of the associations found in the present study were observed with TSH while free T4 was only associated with attention deficit symptoms. Thyroid function can be determined either directly, by measuring the primary thyroid gland product T4 (preferably as free T4), or indirectly, by assessing TSH concentration, which inversely reflects the thyroid hormone concentration sensed by the pituitary. In fact, it is recognized that the indirect approach (serum TSH measurement) offers better sensitivity for detecting thyroid dysfunction than does free T4 testing, for two reasons: serum TSH and free T4 concentrations exhibit an inverse log-linear relationship such that small alterations in free T4 will produce a much larger response in serum TSH;[46] and each individual has a genetically determined free T4 set-point and any mild free T4 excess or deficiency, relative to that individual's free T4 set-point, will be sensed by the pituitary, and cause an amplified, inverse response in TSH secretion.[47] Hence, a thyroid hormone concentration within the laboratory reference range is not necessarily normal for the individual. In the case of subclinical hypothyroidism, serum TSH outside the population-based reference range indicates that serum T3 and T4 are not normal for the individual.[46,48] Nevertheless, despite most of the associations with free T4 not being statistically significant in this study, most are positive (the opposite to TSH), indicating a better cognitive function in children with higher free T4 concentrations.

The major limitation of the present study is its cross-sectional design and hence the lack of repeated measures, which precludes determining the time-window of the effect of TSH (i.e. prenatal, early postnatal or acute at 4 years) and the reversibility of the present findings. Thus, a statistically significant difference does not necessarily mean a causative association. Advantages are that this is the first study in a general population of infants with normal TSH values and neurodevelopment assessment, and that scales affected were a priori formulated, which strengthens the validity of the results. In addition, the consistency of the results between the two cohorts in most of the outcomes also reinforces their validity, despite the small sample size of the Ribera d'Ebre cohort.

In the present study, the highest TSH value was 6·04 mU/l and none of the five children with TSH concentrations higher than 5 mU/l had a diagnosis of thyroid disorder. There is controversy about the reference range of TSH concentrations, but by convention this usually comprises 95% of a reference population.[47,49,50] Most of the upper limits of the serum TSH reference ranges established in different studies are between 3 and 6 mU/l for children and adults.[47,49,50] However, given the high prevalence of mild, subclinical hypothyroidism in the general population, it is likely that the upper limit of the population reference range is biased by the inclusion of people with nondiagnosed dysfunction.[47,50] Subclinical hypothyroidism is a clinical disorder defined by TSH concentrations above the reference range and free T4 and T3 concentrations within the reference range. Although patients with this disorder can be asymptomatic, some patients present cardiovascular or neuropsychiatric alterations.[50–53] Recent reports have suggested that treatment of subclinical hypothyroidism can reduce these effects.[51–53] The British thyroid association recommends treating subclinical hypothyroidism with thyroxine therapy in patients with TSH higher than 10 mU/l.[54]

In the present study, children with higher, but normal TSH concentrations presented lower scores in neurodevelopmental skills, as well as higher risk of attention and hyperactivity/impulsivity symptoms; nevertheless, none of these children is considered as hypothyroid. Further research is required to determine whether these marginal concentrations of TSH in children can still be accepted as 'normal', with regard to infant neurodevelopment and other functions related to thyroid hormones.

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References

1 de Escobar, G.M., Obregon, M.J. & del Rey, F.E. (2004) Maternal thyroid hormones early in pregnancy and fetal brain development. *Best Practice and Research. Clinical Endocrinology and Metabolism*, **18**, 225–248.

2 Chan, S. & Kilby, M.D. (2000) Thyroid hormone and central nervous system development. *Journal of Endocrinology*, **165**, 1–8.

3 Porterfield, S.P. & Hendrich, C.E. (1993) The role of thyroid hormones in prenatal and neonatal neurological development – current perspectives. *Endocrine Reviews*, **14**, 94–106.

4 Anderson, G.W. (2001) Thyroid hormones and the brain. *Frontiers in Neuroendocrinology*, **22**, 1–17.

5 MedlinePlus Medical Encyclopedia. http://www.nlm.nih.gov/medlineplus/ency/article/001193.htm Updated on 20 October 2005 by Nikheel, S., Kolatkar, M.D. Review provided by VeriMed Healthcare Network.

6 Haddow, J.E., Palomaki, G.E., Allan, W.C., Williams, J.R., Knight, G.J., Gagnon, J., O'Heir, C.E., Mitchell, M.L., Hermos, R.J., Waisbren, S.E., Faix, J.D. & Klein, R.Z. (1999) Maternal thyroid deficiency during pregnancy and subsequent neuropsychological development of the child. *New England Journal of Medicine*, **341**, 549–555.

7 Pop, V.J., Kuijpens, J.L., van Baar, A.L., Verkerk, G., van Son, M.M., de Vijlder, J.J., Vulsma, T., Wiersinga, W.M., Drexhage, H.A. & Vader, H.L. (1999) Low maternal free thyroxine concentrations during early

pregnancy are associated with impaired psychomotor development in infancy. *Clinical Endocrinology*, **50**, 149–155.

8 Calvo, R.M., Jauniaux, E., Gulbis, B., Asuncion, M., Gervy, C., Contempre, B. & Morreale de Escobar, G. (2002) Fetal tissues are exposed to biologically relevant free thyroxine concentrations during early phases of development. *Journal of Clinical Endocrinology and Metabolism*, **87**, 1768–1777.

9 Kooistra, L., Laane, C., Vulsma, T., Schellekens, J.M., van der Meere, J.J. & Kalverboer, A.F. (1994) Motor and cognitive development in children with congenital hypothyroidism. *Journal of Pediatrics*, **124**, 903–909.

10 Murphy, G.H., Hulse, J.A., Smith, I. & Grant, D.B. (1990) Congenital hypothyroidism: physiological and psychological factors in early development. *Journal of Child Psychology and Psychiatry*, **31**, 711–725.

11 Rovet, J.F., Ehrlich, R.M. & Sorbara, D.L. (1992) Neurodevelopment in infants and preschool children with congenital hypothyroidism: etiological and treatment factors affecting outcome. *Journal of Pediatric Psychology*, **17**, 187–213.

12 Rovet, J.F. & Ehrlich, R.M. (1995) Long-term effects of 1-thyroxine therapy for congenital hypothyroidism. *Journal of Pediatrics*, **126**, 380–386.

13 Rovet, J. (1999) Congenital hypothyroidism: long-term outcome. *Thyroid*, **9**, 741–748.

14 Weber, G., Mora, S., Prina Cerai, L.M., Siragusa, V., Colombini, J., Medaglini, S., Fornara, C., Locatelli, T., Comi, G. & Chiumello, G. (2000) Cognitive function and neurophysiological evaluation in early-treated hypothyroid children. *Neurological Sciences*, **21**, 307–314.

15 Glorieux, J., Dussault, J.H., Letarte, J., Guyda, H. & Morissette, J. (1983) Preliminary results on the mental development in children with hypothyroidism detected by Quebec Screening Programme. *Journal of Pediatrics*, **102**, 19–22.

16 Glorieux, J., Dussault, J.H., Morissette, J., Desjardins, M., Letarte, J. & Guyda, H. (1985) Follow-up at ages 5 and 7 years on mental development in children with hypothyroidism detected by Quebec Programme. *Journal of Pediatrics*, **107**, 913–915.

17 Tillotson, S.L., Fuggle, P.W., Smith, I., Ades, A.E. & Grant, D.B. (1994) Relation between biochemical severity and intelligence in early treated congenital hypothyroidism: a threshold effect. *British Medical Journal*, **309**, 440–445.

18 New England Congenital Hypothyroidism Collaborative. (1981) Effects of neonatal screening for hypothyroidism: prevention of mental retardation by treatment before clinical manifestations. *Lancet*, **2**, 1095–1098.

19 Bauer, M. & Whybrow, P.C. (2001) Thyroid hormone, neural tissue and mood modulation. *The World Journal of Biological Psychiatry*, **2**, 59–69.

20 Tejani-Butt, S.M., Yang, J. & Kaviani, A. (1993) Time course of altered thyroid states on 5-HT1A receptors and 5-HT uptake sites in rat brain: an autoradiographic analysis. *Neuroendocrinology*, **57**, 1011–1018.

21 Tejani-Butt, S.M. & Yang, J. (1994) A time course of altered thyroid states on the noradrenergic system in rat brain by quantitative autoradiography. *Neuroendocrinology*, **59**, 235–244.

22 Constant, E.L., de Volder, A.G., Ivanoiu, A., Bol, A., Labar, D., Seghers, A., Cosnard, G., Melin, J. & Daumerie, C. (2001) Cerebral blood flow and glucose metabolism in hypothyroidism: a positron emission tomography study. *Journal of Clinical Endocrinology and Metabolism*, **86**, 3864–3870.

23 Yen, P.M. (2001) Physiological and molecular basis of thyroid hormone action. *Physiological Reviews*, **81**, 1097–1142.

24 Madeira, M.D., Sousa, N., Lima-Andrade, M.T., Calheiros, F., Cadete-Leite, A. & Paula-Barbosa, M.M. (1992) Selective vulnerability of the hippocampal pyramidal neurons to hypothyroidism in male and female rats. *Journal of Comparative Neurology*, **322**, 501–518.

25 Rami, A., Patel, A.J. & Rabie, A. (1986) Thyroid hormone and development of the rat hippocampus: morphological alterations in granule and pyramidal cells. *Neuroscience*, **19**, 1217–1226.

26 Gould, E., Allan, M.D. & McEwen, B.S. (1990) Dendritic spine density of adult hippocampal pyramidal cells is sensitive to thyroid hormone. *Brain Research*, **525**, 327–329.

27 Whybrow, P.C. & Bauer, M. (2005) Behavioral and psychiatric aspects of hypothyroidism. In: Braverman, L.E., Utiger, R.D. eds. *The Thyroid*, 9th edn. Lippincott Williams & Wilkins, Philadelphia, 842–849.

28 Whybrow, P.C. & Bauer, M. (2005) Behavioral and psychiatric aspects of thyroxicosis. In: Braverman, L.E., Utiger, R.D. eds. *The Thyroid*, 9th edn. Lippincott Williams & Wilkins, Philadelphia, 644–650.

29 Dugbartey, A.T. (1998) Neurocognitive aspects of hypothyroidism. *Archives of International Medicine*, **158**, 1413–1418.

30 Polk, S., Sunyer, J., Munoz-Ortiz, L., Barnes, M., Torrent, M., Figueroa, C., Harris, J., Vall, O., Anto, J.M. & Cullinan, P. (2004) A prospective study of Fel d1 and Der p1 exposure in infancy and childhood wheezing. *American Journal of Respiratory and Critical Care Medicine*, **170**, 273–278.

31 Ribas-Fito, N., Cardo, E., Sala, M., de Muga, E., Mazon, C., Verdu, A., Kogevinas, M., Grimalt, J.O. & Sunyer, J. (2003) Breastfeeding, exposure to organochlorine compounds, and neurodevelopment in infants. *Pediatrics*, **111**, e580–e585.

32 Ribas-Fito, N., Torrent, M., Carrizo, D., Muñoz-Ortiz, L., Julvez, J., Grimalt, J.O. & Sunyer, J. (2006) In utero exposure to background concentrations of DDT and cognitive functioning among preschoolers. *American Journal of Epidemiology*, **164**, 955–962.

33 McCarthy, D. (1972) *Manual for the McCarthy Scales of Children's Abilities*. Psychological Corp, New York.

34 American Psychiatric Association (1994) *Diagnostic and Statistical Manual of Mental Disorders (DSM-IV)*, 4th edn. American Psychiatric Association, Washington, DC.

35 Grimalt, J.O., Sunyer, J., Moreno, V., Amaral, O.C., Sala, M., Rosell, A., Anto, J.M. & Albaiges, J. (1994) Risk excess of soft-tissue sarcoma and thyroid cancer in a community exposed to airborne organochlorinated compound mixtures with a high hexachlorobenzene content. *International Journal of Cancer*, **56**, 200–203.

36 Sala, M., Sunyer, J., Herrero, C., To-Figueras, J. & Grimalt, J.O. (2001) Association between serum concentrations of hexachlorobenzene and polychlorobiphenyls with thyroid hormone and liver enzymes in a sample of the general population. *Occupational Environmental Medicine*, **58**, 172–177.

37 Utiger, R.D. (1999) Editorial: Maternal hypothyroidism and fetal development. *New England Journal of Medicine*, **341**, 601–602.

38 Vermiglio, F., Lo Presti, V.P., Moleti, M., Sidoti, M., Tortorella, G., Scaffidi, G., Castagna, M.G., Mattina, F., Violi, M.A., Crisa, A., Artemisia, A. & Trimarchi, F. (2004) Attention deficit and hyperactivity disorders in the offspring of mothers exposed to mild-moderate iodine deficiency: a possible novel iodine deficiency disorder in developed countries. *Journal of Clinical Endocrinology and Metabolism*, **89**, 6054–6060.

39 Stein, M.A., Weiss, R.E. & Refetoff, S. (1995) Neurocognitive characteristics of individuals with resistance to thyroid hormone: comparisons with individuals with attention-deficit hyperactivity disorder. *Journal of Developmental and Behavioral Pediatrics*, **16**, 406–411.

40 Ganguli, M., Burmeister, L.A., Seaberg, E.C., Belle, S. & DeKosky, S.T. (1996) Association between dementia and elevated TSH: a community-based study. *Biological Psychiatry*, **40**, 714–725.

41 Stein, M.A. & Weiss, R.E. (2003) Thyroid function tests and neurocognitive functioning in children referred for attention deficit/hyperactivity disorder. *Psychoneuroendocrinology*, **28**, 304–316.

42 Weiss, R.E., Stein, M.A., Trommer, B. & Refetoff, S. (1993) Attention-deficit hyperactivity disorder and thyroid function. *The Journal of Pediatrics*, **123**, 539–545.

43 Dorn, L.D. (1997) Baseline thyroid hormones in depressed and non-depressed pre- and early-pubertal boys and girls. *Journal of Psychiatric Research*, **31**, 555–567.

44 Wahlin, A., Wahlin, T.B., Small, B.J. & Backman, L. (1998) Influences of thyroid stimulating hormone on cognitive functioning in very old age. *The Journals of Gerontology: Psychological Sciences and Social Sciences*, **53**, 234–239.

45 Wahlin, A., Bunce, D. & Wahlin, T.B. (2005) Longitudinal evidence of the impact of normal thyroid stimulating hormone variations on cognitive functioning in very old age. *Psychoneuroendocrinology*, **30**, 625–637.

46 Andersen, S., Pedersen, K.M., Bruun, N.H. & Laurberg, P. (2002) Narrow individual variations in serum T(4) and T(3) in normal subjects: a clue to the understanding of subclinical thyroid disease. *Journal of Clinical Endocrinology and Metabolism*, **87**, 1068–1072.

47 Demers, L.M. & Spencer, C.A. (2002) *Laboratory Medicine Practice Guidelines*. Laboratory support for the Diagnosis and Monitoring of Thyroid Disease, National Academy of Clinical Biochemistry (NACB). Available at: http://www.nacb.org/lmpg/thyroid_LMPG_Word.stm (accessed 25 May 2006).

48 Meier, C.A., Maisey, M.N., Lowry, A., Muller, J. & Smith, M.A. (1993) Interindividual differences in the pituitary-thyroid axis influence the interpretation of thyroid function tests. *Clinical Endocrinology*, **39**, 101–107.

49 Elmlinger, M.W., Kuhnel, W., Lambrecht, H.G. & Ranke, M.B. (2001) Reference intervals from birth to adulthood for serum thyroxine (T4), triiodothyronine (T3), free T3, free T4, thyroxine binding globulin (TBG) and thyrotropin (TSH). *Clinical Chemistry and Laboratory Medicine*, **39**, 973–979.

50 Surks, M.I., Ortiz, E., Daniels, G.H., Sawin, C.T., Col, N.F., Cobin, R.H., Franklyn, J.A., Hershman, J.M., Burman, K.D., Denke, M.A., Gorman, C., Cooper, R.S. & Weissman, N.J. (2004) Subclinical thyroid disease: scientific review and guidelines for diagnosis and management. *JAMA*, **291**, 228–238.

51 Baldini, I.M., Vita, A., Mauri, M.C., Amodei, V., Carrisi, M., Bravin, S. & Cantalamessa, L. (1997) Psychopathological and cognitive features in subclinical hypothyroidism. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, **21**, 925–935.

52 McDermott, M.T. & Ridgway, E.C. (2001) Subclinical hypothyroidism is mild thyroid failure and should be treated. *Journal of Clinical Endocrinology and Metabolism*, **86**, 4585–4590.

53 Razvi, S., Ingoe, L., Keeka, G., Oates, C., McMillan, C. & Weaverm, J.U. (2007) The beneficial effect of 1-thyroxine on cardiovascular risk factors, endothelial function and quality of life in subclinical hypothyroidism: randomised, crossover trial. *Journal of Clinical Endocrinology and Metabolism*, in press.

54 British Thyroid Association. (2006) *UK Guidelines for Thyroid Function Tests*, July. Available at: http://www.british-thyroid-association.org/TFT_guideline_final_version_July_2006.pdf (accessed 24 July 2006).