Thyroid

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In this year's overview of thyroid research the beauty of science was revealed in one paper from the far North of Europe, from Iceland. In the darkness of hypothesis-free genome-wide association studies, where nowadays over 300,000 common genetic variants can be tested for an association with a given phenotype, the colleagues from Iceland found two luminous variants which increase the risk of thyroid differentiated cancer (papillary and follicular PTC and FTC, respectively). These two variants are located close to two transcription factor genes which are known for their role in thyroid growth, proliferation and differentiation, i.e. FOXE1 (formerly TTF2) and NKX2.1 (formerly TTF1). Although the search for the underlying molecular impact of these variants on gene function, either gain or loss, needs to be determined now, it is an illuminating moment when a hypothesis-free experiment, which is based on the genotyping of more than 300,000 SNPs in 15,000 individuals, leads to such a conceptionally consistent result.

Besides this wonderful paper there were more than 3,000 publications related to thyroid research in the past 12 months, 18 are summarized in this chapter. Since this chapter will be the last one in the series of my contribution to the *Yearbook*, I will take the opportunity to apologize not only to the authors of the 2,982 papers not mentioned this year but also to the more than 15,000 publications of the last 5 years for not having referred to their work in these thyroid chapters – it was a very subjective choice!

Concept of the year: thyroid cancer meets thyroid development

Common variants on 9q22.33 and 14q13.3 predispose to thyroid cancer in European populations

Gudmundsson J, Sulem P, Gudbjartsson DF, Jonasson JG, Sigurdsson A, Bergthorsson JT, He H, Blondal T, Geller F, Jakobsdottir M, Magnusdottir DN, Matthiasdottir S, Stacey SN, Skarphedinsson OB, Helgadottir H, Li W, Nagy R, Aguillo E, Faure E, Prats E, Saez B, Martinez M, Eyjolfsson GI, Bjornsdottir US, Holm H, Kristjansson K, Frigge ML, Kristvinsson H, Gulcher JR, Jonsson T, Rafnar T, Hjartarsson H, Mayordomo JI, de la Chapelle A, Hrafnkelsson J, Thorsteinsdottir U, Kong A, Stefansson K
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Nat Genet 2009;41:460–464

Background: Thyroid carcinoma is the most common endocrine malignancy. It has been estimated that the risk of thyroid cancer has a greater genetic component than the risk of any other cancer.

Methods: In order to search for sequence variants conferring risk of thyroid cancer, a genome-wide association study in 192 and 37,196 Icelandic cases and controls, respectively, was conducted followed by a replication study in individuals of European descent.

Results: The data show that two common variants, located on 9q22.33 and 14q13.3, are associated with the disease. Overall, the strongest association signals were observed for rs965513 on 9q22.33 (OR = 1.75; $p = 1.7 \times 10^{-27}$) and rs944289 on 14q13.3 (OR = 1.37; $p = 2.0 \times 10^{-9}$). The genes nearest to the 9q22.33 locus are FOXE1 (TTF2) and NKX2-1 (TTF1), which are among the genes located at the 14q13.3 locus. Both variants contribute to an increased risk of both papillary and follicular thyroid cancer. Approximately 3.7% of individuals are homozygous for both variants, and their estimated risk of thyroid cancer is 5.7-fold greater than that of noncarriers. In a study on a large sample set from the general population, both risk alleles are associated with low concentrations of thyroid-stimulating hormone (TSH), and the 9q22.33 allele is associated with low concentration of thyroxine (T_4) and high concentration of triiodothyronine (T_3).

Conclusion: Sequence variants in two genes that are known for their critical role in thyroid development are associated with an increased risk for thyroid carcinoma.

The genetic contribution to the pathogenesis of medullary thyroid cancer (MTC), frequently as part of MEN syndrome, was among the first clinically relevant genetic findings in endocrinology. Nowadays, prophylactic thyroidectomy after the molecular diagnosis of activating RET gene mutations helps to prevent cancer-related death in thousands of patients worldwide. The differentiated thyroid cancer types like papillary (PTC) and follicular (FTC) thyroid cancer are less likely to be associated with a genetic predisposition as MTC but still have a high familial risk in first-degree relatives. How this risk is inherited has not been determined so far. This paper of the year in the thyroid chapter used the power of the Iceland genotyping project to unravel the common genetic variants that are associated with an increased risk for PTC and FTC. Compared to the common susceptibility variants recently discovered for complex diseases, thyroid cancer may not be as complex. Out of 304,083 single nucleotide polymorphisms (SNP), only 2 (!) were found to be associated with an increased risk. The incidence of thyroid cancer, i.e. 4.6 and 12.1 in 100,000 males and females, respectively, is 5.7-fold greater in individuals with double homozygosity for the two predisposing SNP variants AA and TT. This result does not reach a clinical relevance as the finding of an activating mutation in the RET gene which renders the carrier at risk to develop MTC is 100%. The double homozygous SNP variant carriers will most probably *not* develop PTC or FTC: only 47 of 100,000 will have cancer and therefore the data have no clinical relevance in terms of prophylactic thyroidectomy!

However, the beauty of the study is the nature of the genes in which the identified variants are located: both genes, FOXE1 (also named initially TTF2) and NKX2.1 (initially named TTF1) are well known for their role in thyroid development and as candidate genes for syndromic thyroid dysgenesis. From the extremely large number of possible genes related to cell growth and cell cycle regulation which could potentially be linked to cancer susceptibility, only these two genes, which are established key transcription factors for thyroid cell growth and differentiation [1], were located closely to the risk SNPs. These data might tell us that the transcriptional regulation of embryogenesis of the thyroid gland is closely related to the transformation process of differentiated thyroid follicular cells into less differentiated cancer cells. The question is how the two variants in FOXE1 and NKX2.1 increase the thyroid cancer risk. In principle, SNPs outside the coding region of genes can either increase or decrease the function of the gene by modification of transcription and translation. Both transcription factor genes are involved in thyroid growth and differentiation. Therefore, loss or gain of function could contribute to tumorigenesis. If loss of function caused the increased cancer risk, patients with CH and loss of function in FOXE1 and NKX2.1 would harbor a so far unrecognized thyroid cancer risk.

The following study, which was published before the Iceland thyroid cancer SNP data, already gives some answers to these critical questions.

A germline mutation (A339V) in thyroid transcription factor-1 (TTF-1/NKX2.1) in patients with multinodular goiter and papillary thyroid carcinoma

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J Natl Cancer Inst 2009;101:162–175

Background: To determine the molecular basis of the risk of papillary thyroid carcinoma (PTC) among patients with multinodular goiter (MNG), the authors hypothesized that mutations in the gene that encode NKX2.1 (TTF-1) are a genetic determinant of MNG/PTC predisposition.

Methods: 20 unrelated PTC patients with a history of MNG (MNG/PTC), 284 PTC patients without a history of MNG (PTC), and 349 healthy control subjects were screened for germline mutation(s) in TTF-1/NKX2.1 by sequencing of amplified DNA from blood. The effects of the mutation on the growth and differentiation of thyroid cells were demonstrated by ectopic expression of wild-type (WT) and mutant proteins in PCCL3 normal rat thyroid cells, followed by tests of cell proliferation, activation of cell growth pathways, and transcription of TTF-1 target genes.

Results: A missense mutation (1016C>T) was identified in TTF-1/NKX2.1 that led to a mutant TTF-1 protein (A339V) in 4 of the 20 MNG/PTC patients (20%). These patients developed substantially more advanced tumors than MNG/PTC or PTC patients without the mutation ($p = 0.022$, Fisher exact test). Notably, this germline mutation was dominantly inherited in two families, with some members bearing the mutation affected with MNG, associated with either PTC or colon cancer. The mutation encoding the A339V substitution was not found among the 349 healthy control subjects nor among the 284 PTC patients who had no history of MNG. Overexpression of A339V TTF-1 in PCCL3 cells, as compared with overexpression of WT TTF-1, was associated with increased cell proliferation including thyrotropin-independent growth (average A339V proliferation rate = 134.27%, WT rate = 104.43%, difference = 34.3%, 95% CI = 12.0–47.7%, $p = 0.010$), enhanced STAT3 activation, and impaired transcription of the thyroid-specific genes Tg, TSH-R, and Pax-8.

Conclusion: This is the first germline mutation identified in MNG/PTC patients. It could contribute to predisposition for MNG and/or PTC and to the pathogenesis of PTC. Joined with the Iceland data, one might expect that the SNPs in the NKX2.1 gene that increase the risk for thyroid cancer more likely lead to an increased expression of the gene.

Before knowing the results from Iceland, the colleagues from Hong Kong followed a candidate concept for the genetics of PTC and speculated that those PTC that develop in goiter glands would more likely be linked to a genetic germline predisposition in a gene of thyroid growth regulation like NKX2.1. They identified a likely gain of function germline mutation in a rather high number of patients with PTC and goiter. According to these data, it seems likely that activation of the NKX2.1 gene leads to increased proliferation and dedifferentiation, and might cause thyroid cancer. These will pave the way for further experiments that will focus on the functional relevance of the NKX2.1 gene-associated SNPs in the Iceland study. For pediatric patients with loss of function mutations in NKX2.1, who are affected by thyroid dysfunction and movement defects, these data represent a relief; it is unlikely that their loss of NKX2.1 function renders them susceptible for thyroid cancer. In the following sections the two themes of the paper of the year, thyroid cancer and thyroid development, will be dwelled upon and interesting papers focusing on thyroid childhood cancer and thyroid development will be discussed.

Clinical concepts in childhood thyroid cancer

Ultrasound screening for thyroid carcinoma in childhood cancer survivors: a case series

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J Clin Endocrinol Metab 2008;93:4840–4843

Background: Childhood cancers are at risk for developing late-onset complications of cancer therapy, including thyroid carcinoma in childhood cancer survivors. How to screen for thyroid cancer in these patients is a matter of debate.

Methods: A total of 129 subjects who had received radiotherapy to the head, neck, or upper thorax for a pediatric cancer were studied in the setting of a long-term follow-up unit. Thyroid ultrasound usually began 5 years after radiotherapy and was repeated every third year, if negative. Median follow-up time since childhood cancer diagnosis was 15.8 years (range 6.1–34.8). Solid thyroid nodules were found in 35 patients. Fine-needle aspiration was performed in 19 patients, of which 14 had nodules >1 cm. The main outcome measure was the finding of not palpable thyroid cancers.

Results: Cytological examination of specimens diagnosed papillary carcinoma in 5 patients who underwent surgery. The cytological diagnosis of papillary thyroid carcinoma was confirmed in all cases by histological examination. Notably, only 2 of these patients had palpable nodules; the other 3 were <1 cm and were detected only by ultrasound. However, histological examination showed nodal metastases in 2 of these.

Conclusion: Although ultrasound screening for thyroid cancer in the general population is not cost-effective and could lead to unnecessary surgery, due to false positives, they believe that in childhood cancer survivors who received radiotherapy involving the head, neck, or upper thorax, it would be worthwhile.

With the increasing use of thyroid ultrasound as a screening tool for thyroid nodules, a new cohort of 'potential' patients is being diagnosed without a clear concept of how to treat and how to follow these children with 'thyroid incidentalomas'. Population-based nodule screening by palpation or by ultrasound revealed a prevalence of 0.6% single nodules in children and adolescents [2]. Even though this is by far less than in the adult population [3], we do not want to biopsy 6 in 1,000 children. Because few studies with preselected patients claimed a high rate of malignancy in childhood thyroid nodules, the diagnosis of a nodule is causing for the most part severe concern and fear in the patients and their families. However, the reported malignancy rate of up to 20% of childhood thyroid nodules is not affirmed by population-based calculations of reported thyroid cancer cases in childhood and a far lower rate can be expected. Moreover, with the advancement of ultrasound techniques, more and more smaller nodules are being detected and their malignancy potential is not clear. The available data does not justify the screening of all children for thyroid nodules but some groups of patients with an increased risk of thyroid malignancy should be followed more carefully. This paper presents data about a group of children with primary malignancies and head and neck irradiation as part of their treatment. Even in this thyroid cancer-prone group of patients, only a small number of cases with thyroid cancer was identified by the ultrasound screening program (5 of 129). These results are different from a recent large thyroid FNA study which showed that about 30% of patients having thyroid nodules with a cytological diagnosis of follicular neoplasm had histologically confirmed malignancy [4]. Moreover, the identified secondary thyroid cancers did not develop distant metastases, and belong therefore to the group of PTC with a very good prognosis given that therapy with radioiodine is performed. The histologically diagnosed thyroid cancers were adequately prediagnosed by fine-needle aspiration. In the particular group of patients with primary cancer and irradiation an ultrasound screening for secondary thyroid cancer seems to be useful to identify PTCs that have a good prognosis before they develop distant metastasis.

Three of the 5 diagnosed thyroid cancers were <1 cm and therefore fulfilled the criteria of 'microcarcinoma'. These very small tumors are difficult to diagnose by FNA and their clinical course is very difficult to predict. The following nice review was published with a summary of available data for microcarcinoma, and it turns out that the estimated risk for a less favorable course with higher recurrence risk is related mainly to a younger age <45 years (but childhood was not covered), a lymph node infiltration and a clinical sign of palpability. However, we need to keep in mind that about half of the ultrasound-detected nodules >1.5 cm were not palpated by 21st century endocrinologists [5]. Since the data are generated mainly for adults, microcarcinomas in children will further lead to uncertainty, but in general microcarcinomas have a more benign course.

Thyroid papillary microcarcinoma: a descriptive and meta-analysis study

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Eur J Endocrinol 2008;159:659–673

Background: Microcarcinomas of the thyroid are frequent and prediction of the course of further growth and malignancy of the tumors are of utmost clinical importance.

Methods/Results: The authors review anatomical, clinical characteristics and prevalence of thyroid microcarcinoma. Diagnostic procedures and risk factors of aggressiveness at diagnosis and during follow-up are also covered. The possible clinical, pathologic and therapeutic risk factors are analyzed by meta-analysis study. Treatment procedures by different authors and guidelines suggested by societies are reported.

Conclusion: Papillary thyroid microcarcinoma has in general a benign course and sophisticated diagnostics and aggressive treatment appear unnecessary.

Pathways revised: the right surrounding for normal thyroid development

For normal thyroid development, precursor cells require an intrinsic regulatory cascade, of which we know three transcription factors, e.g. FOXE1, NKX2.1 and PAX8, which are affected by loss of function mutations in patients with thyroid dysgenesis. However, the developing thyroid precursor cells need to be embedded in a growth and differentiation-promoting environment, which is defined by a variety of different growth factors. In the last year, three papers, which are summarized below, described pathways which are involved in these external signals. These signals are required for the thyroid precursor to find its way to the lateral neck position and to grow appropriately. Members of these growth factor cascades are already known from areas of other organ developments and are involved in complex genetic diseases like the CATCH 22 syndrome, a disease that rarely involves thyroid dysgenesis like hemithyroid. The major candidate gene within the Catch22-deleted region in Tbx1 and Tbx1 knockout mouse resembles the asymmetric hypoplastic thyroid phenotype of some Catch22 patients. Altogether, we have learned a lot about the complexity of mesodermal signaling cascades that are involved in thyroid development, but diseases of such factors are rare and result in much more complex syndromes than thyroid dysgenesis itself. We still need to learn more about the critical steps of thyroid development to find the key for the pathogenesis of congenital hypothyroidism. The findings are summarized in figure 1.

Fig. 1. Summary of signal pathways that are involved in thyroid growth and proper localization during early steps of embryogenesis. In addition to transcription factors that are expressed within the thyroid primordium itself, several regulatory factors were described in the last years, which are involved in the 'external' growth promotion and localization of the primordium. The first of these factors was Tbx1 – located within the deleted region of Catch22 patients. Tbx1 knockout mice have a misplaced single-lobed thyroid gland, as it was reported for few patients with Catch22 deletions. The three papers summarized have now described the Tbx1 signal cascade in more detail. Lania et al. demonstrated that within the Tbx1-positive mesodermal cells, Fgf8 is one downstream effector because Tbx1 cell-specific Fgf8 knock-out revealed the same asymmetric and hypoplastic thyroid phenotype as Tbx1 knockout itself. Most likely, but not conclusively proven, one target of the Fgf8 signal are neural crest cells in the surrounding of Tbx1-positive cells because in these cells several components of the Fgf-receptor signal cascade are involved in thyroid shaping as well (as shown by Kameda et al. for FRS2 α and by Newbern et al. for Erk1/2). How these signals are transferred to the thyroid primordium and how the mesodermal cascade is initiated remains unclear.

Early thyroid development requires a Tbx1-Fgf8 pathway

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Dev Biol 2009;328:109–117

Background: The thyroid develops within the pharyngeal apparatus from endodermally-derived cells. The many derivatives of the pharyngeal apparatus develop at similar times and sometimes from common cell types, explaining why many syndromic disorders express multiple birth defects affecting different structures that share a common pharyngeal origin. Thus, different derivatives may share common genetic networks during their development. Tbx1, the major gene associated with DiGeorge syndrome, is a key player in the global development of the pharyngeal apparatus, being required for virtually all its derivatives, including the thyroid.

Methods/Results: The authors show by studying a variety of different knockout mouse lines that Tbx1 regulates the size of the early thyroid primordium through its expression in the adjacent mesoderm. Because Tbx1 regulates the expression of Fgf8 in the mesoderm, they postulated that Fgf8 mediates critical Tbx1-dependent interactions between mesodermal cells and endodermal thyrocyte progenitors. Conditional ablation of Fgf8 in Tbx1-expressing cells caused an early thyroid phenotype similar to that of Tbx1 mutant mice. In addition, expression of an Fgf8 cDNA in the Tbx1 domain rescued the early size defect of the thyroid primordium in Tbx1 mutants.

Conclusion: The authors have established that a Tbx1→Fgf8 pathway in the pharyngeal mesoderm is a key size regulator of mammalian thyroid.

Mouse and human phenotypes indicate a critical conserved role for ERK2 signaling in neural crest development

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Proc Natl Acad Sci USA 2008;105:17115–17120

Background: Disrupted ERK1/2 (MAPK3/MAPK1) MAPK signaling has been associated with several developmental syndromes in humans; however, mutations in ERK1 or ERK2 have not been described.

Results: The authors demonstrate haplo-insufficient ERK2 expression in patients with a novel approximately 1 Mb microdeletion in distal 22q11.2, a region that includes ERK2. These patients exhibit conotruncal and craniofacial anomalies that arise from perturbation of neural crest development and exhibit defects comparable to the DiGeorge syndrome spectrum. Remarkably, these defects are replicated in mice by conditional inactivation of ERK2 in the developing neural crest. Inactivation of upstream elements of the ERK cascade (B-Raf and C-Raf, MEK1 and MEK2) or a downstream effector, the transcription factor serum response factor resulted in analogous developmental defects including thyroid malpositioning.

Conclusion: The findings demonstrate that mammalian neural crest development is critically dependent on a RAF/MEK/ERK/serum response factor signaling pathway and suggest that neural crest autonomous ERK2 signaling is important for normal thyroid development.

FRS2 α is required for the separation, migration, and survival of pharyngeal-endoderm derived organs including thyroid, ultimobranchial body, parathyroid, and thymus

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Background: The docking protein FRS2 α plays an important role in fibroblast growth factor (FGF)-induced intracellular signal transduction by linking FGF receptors (FGFRs) to a variety of intracellular signaling pathways. Given the potential role of Fgf10 and Fgf8 in normal thyroid development, FRS2 α might also be involved in normal thyroid organogenesis.

Results: In FRS2 α (2F/2F) mutant mice at embryonic day (E)18.5, in which the Shp2-binding sites of FRS2 α were disrupted, the thyroid glands were aplastic or hypoplastic. C cells were absent or present in low numbers and rarely formed a compact mass of cells. Parathyroid glands were mostly connected to thymus tissues. At E10.5, the formations of pharyngeal pouches and thyroid primordium were normally initiated in the mutant mice. At E11.5–E12.5, the thyroid primordium of wild-type embryos was located close to the aortic sac, and the epithelial buds of pharyngeal-derived organs, including the parathyroid gland, thymus and ultimobranchial body, were separated from the epithelium and began to migrate to their final destinations. In the FRS2 α (2F/2F) mutants, however, the thyroid primordium became hypoplastic and the pharyngeal-derived organ primordia remained affiliated with the pharyngeal epithelium. At these stages, organ-specific differentiation markers (i.e., Nkx2-1/TTF1 for the thyroid lobe and ultimobranchial body; Pax8 for the thyroid lobe; parathormone (PTH), chromogranin A, P75(NTR), and S100 protein for the parathyroid gland, and p63 for the thymus) were normally expressed in the mutant tissues.

Conclusion: The separation, migration, and survival of the pharyngeal organs including the thyroid were impaired in the FRS2 α (2F/2F) mutants.

Thyrotropin-independent induction of thyroid endoderm from embryonic stem cells by activin A

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Endocrinology 2009;150:1970–1975

Background: Studies with embryonic stem cells can further unravel relevant factors for early embryogenesis. To model the differentiation of thyroid epithelial cells, the authors examined embryoid bodies derived from undifferentiated murine embryonic stem cells treated with activin A to induce endoderm differentiation, the germ layer from which thyroid cells occur.

Methods: Resulting endodermal cells were then further exposed to TSH and/or IGF-I for up to 21 days.

Results: Oct-4 and REX1 expression, required to sustain stem cell self-renewal and pluripotency, were appropriately downregulated, whereas GATA-4, and α -fetoprotein, both endodermal-specific markers, increased as the embryonic stem cells were exposed to activin A. By day 5 culture, TSH receptor (TSHR) and sodium iodide symporter (NIS) gene and protein expression were markedly induced. Cells isolated by the fluorescence-activated cell sorter simultaneously expressed not only TSHR and NIS proteins but also PAX8 mRNA, an expression pattern unique to thyroid cells and expected in committed thyroid progenitor cells. Such expression continued until day 21 with no influence seen by the addition of TSH or IGF-I.

Conclusion: The sequence of gene expression changes observed in these experiments demonstrated the emergence of definitive thyroid endoderm. The activin A induction of thyroid-specific markers, NIS and TSHR, occurred in the absence of TSH stimulation, and, therefore, the emergence of thyroid endoderm in vitro parallels the emergence of thyroid cells in TSHR knockout mice. Activin A is clearly a major regulator of thyroid endoderm.

This last paper in the series of relevant factors in thyroid development is unique in that a reverse approach is taken. The authors are pioneers in establishing the experimental settings to generate thyroid cells from embryonic stem cells. Applying this strategy, growth factors can be tested in vitro for their contribution to thyroid differentiation and commitment. Employing this experimental setting, the authors demonstrate that activin A, a growth factor known to play a pivotal role in the organogenesis of other endodermal areas, seems to have the potential for thyroid determination. The overall concept of early organ determination within a developmental field like the anterior endoderm is based on the assumption that a gradient of growth factors defines a particular concentration that will direct the uncommitted cell to start an organ-specific program. For the thyroid gland, one of the early growth factors could be activin A.

Since the introduction of newborn screening for congenital hypothyroidism the key question remains if an early postnatal treatment with thyroid hormone can enable a completely normal cognitive outcome and if the fetal hypothyroidism itself does cause a noncompensable defect of mental development. The early reports of final cognitive outcome in young adult patients diagnosed in screening programs have shown a remarkably normal outcome but still with a gap of 8 IQ points less compared to appropriate control groups [6, 7]. However, in these cohorts, treatment started later, within the second week of life and consisted of doses $<10 \mu\text{g/kg}$. Now the first cohort that was treated very early and with a high initial dose is reported from Switzerland.

Children with congenital hypothyroidism: long-term intellectual outcome after early high-dose treatment

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Pediatr Res 2009;65:242–248

Background: Outcome studies in patients with congenital hypothyroidism (CH) diagnosed in newborn screening programs revealed a significant 6- to 10-point difference in IQ. The question remains if a very early and high-dose treatment can close this gap in cognitive development.

Methods: 63 prospectively followed children with CH were assessed at age of 14 years with the Wechsler Intelligence Scale for Children-Revised and compared with 175 healthy controls. Median age at onset of treatment was 9 days (range 5–18) and median starting dose of levothyroxine (L-T_4) was $14.7 \mu\text{g/kg/day}$ (range 9.9–23.6).

Results: Full-scale intelligence quotient (IQ) was significantly lower than in controls after adjustment for socioeconomic status (SES) and gender (101.7 vs. 111.4; $p < 0.0001$). Children with athyreosis had a lower performance IQ than those with dysgenesis (adjusted difference 7.6 IQ scores, $p < 0.05$). Lower initial thyroxine (T_4) levels correlated with poorer IQ ($r = 0.27$, $p = 0.04$). Lower SES was associated with poorer IQ, in particular in children with CH (interaction, $p = 0.03$). Treatment during childhood was not related to IQ at age 14 years.

Conclusion: Adolescents with CH manifest IQ deficits when compared with their peers despite early high-dose treatment and optimal substitution therapy throughout childhood.

Thus, the gap still exists! Obviously these data are a drawback for the hope of completely normal IQ in patients with congenital hypothyroidism. But, some important questions still need to be answered. As it was shown that congenital hypothyroidism can be part of more complex endocrine developmental diseases like pseudohypoparathyroidism and the more recently described gene mutations that are important for thyroid as well as brain development (FOXE1 and NKX2.1, see above), a more detailed analysis of the cases which did not reach satisfactory outcome, and which might have decreased the overall IQ level of the patient cohort, are mandatory. Based on these findings of associated brain defects, an alternative explanation for the IQ gap in patients with congenital hypothyroidism is that subgroups of patients are affected by associated and not directly causally related genetic brain defects, and that these deficits persist independently of the thyroid hormone treatment efforts. A more thorough search for such defects and an exclusion of these cases from the outcome cohorts might end up with IQ levels showing no gap.

Early and high-dose treatments are only part of the strategy, which this study addresses. Quite as important is the maintenance throughout childhood of normal TSH.

The principal message to parents with a newborn with congenital hypothyroidism is that for most children cognitive development will be within the normal range, provided that therapy is given using a full dose and TSH is monitored often enough.

Although the gap still exists, the question whether a higher dose is beneficial compared to a lower dose is not further answered by this new publication. The evidence for the use of a higher dose is still low, and the only existing prospective randomized study, comparing 37 and $50 \mu\text{g/day}$ is in favor of a higher dose [8]. Nevertheless, the next report from the Cochrane Collaboration this year shows our limits of evidence and makes it obvious that the evidence for the higher dose is not strong enough to be officially recommended.

High versus low dose of initial thyroid hormone replacement for congenital hypothyroidism

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Cochrane Database Syst Rev 2009;CD006972

Background: The authors aimed to determine the evidence for effects of high versus low dose of initial thyroid hormone replacement for congenital hypothyroidism.

Methods: Randomized controlled trials were identified by searching The Cochrane Library, MEDLINE and EMBASE and reference lists of published papers. Randomized controlled clinical trials investigating the effects of high versus low dose of initial thyroid hormone replacement for congenital hypothyroidism were included. Both authors independently selected trials, assessed risk of bias and extracted data.

Results: The initial search identified 1,014 records, which identified 13 publications for further examination. After screening the full text of the 13 selected papers, only one study evaluating 47 babies finally met the inclusion criteria. Using the same cohort at two different time periods, the study investigated the effects of high versus low dose thyroid hormone replacement in relation to (1) time taken to achieve euthyroid status and (2) neurodevelopmental outcome. The study reported that a high dose is more effective in rising serum thyroxine and free thyroxine concentrations to the target range and earlier normalization of thyroid-stimulating hormone compared to a lower dose. Similarly, full-scale intelligence quotient (IQ) was noted to be significantly higher in children who received the high dose compared to the lower dose. However, the verbal IQ and performance IQ were similar in both groups. Growth and adverse effects were not reported in the included trial.

Conclusion: There is currently only one randomized controlled trial evaluating the effects of high versus low dose of initial thyroid hormone replacement for CHT. There is inadequate evidence to suggest that a high dose is more beneficial compared to a low dose initial thyroid hormone replacement in the treatment of CHT.

What should we do? First, we need more studies which compare the outcome in relation to the initial treatment dose and large enough to define subgroups with different forms of the disease including the exemption of genetic syndromic defects. Due to the low incidence of the disease, such studies can only be conducted in a multicentered, international effort. So far, almost all outcome studies of congenital hypothyroidism come from single centers and patient groups are too small to allow significant conclusions. Until then, we should continue with the actual recommendations since the available studies are in favor of a better outcome with the higher dosage and no relevant side effects of the higher dose have been reported. It will take many years and births of thousands of children with congenital hypothyroidism until we will have enough data for evidence-based decisions. It would be unethical to treat until then with a lower dose.

Concepts revised: course and causes of 'subclinical' thyroid dysfunction

Neonatal thyroid function in Seveso 25 years after maternal exposure to dioxin

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PLoS Med 2008;5:e161

Background: Neonatal hypothyroidism has been associated in animal models with maternal exposure to several environmental like 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD), a persistent and widespread toxic environmental contaminant.

Methods/Results: Between 1994 and 2005, in individuals exposed to TCDD after the 1976 Seveso accident, we conducted: (i) a residence-based population study on 1,014 children born to the 1,772 women of reproductive age in the most contaminated zones (A, very high contamination; B, high contamination), and 1,772 age-matched women from the surrounding noncontaminated area (reference); (ii) a bio-marker study on 51 mother-child pairs for whom recent maternal plasma dioxin measurements were available. Neonatal blood thyroid-stimulating hormone (b-TSH) was measured on all children. We performed crude and multivariate analyses adjusting for gender, birth weight, birth order, maternal age, hospital, and type of delivery. Mean neonatal b-TSH was 0.98 $\mu\text{U/ml}$ (95% CI 0.90–1.08) in the reference area ($n = 533$), 1.35 $\mu\text{U/ml}$ (95% CI 1.22–1.49) in zone B ($n = 425$), and 1.66 $\mu\text{U/ml}$ (95% CI 1.19–2.31) in zone A ($n = 56$) ($p < 0.001$). The proportion of children with b-TSH $> 5 \mu\text{U/ml}$ was 2.8% in the reference area, 4.9% in zone B, and 16.1% in zone A ($p < 0.001$). Neonatal b-TSH was correlated with current maternal plasma TCDD ($n = 51$, $\beta = 0.47$, $p < 0.001$) and plasma toxic equivalents of coplanar dioxin-like compounds ($n = 51$, $\beta = 0.45$, $p = 0.005$).

Conclusion: The data indicate that environmental contaminants such as dioxins have a long-lasting capability to modify neonatal thyroid function after the initial exposure.

Almost all available epidemiological studies failed to demonstrate a significant impact of environmental factors on the incidence of congenital hypothyroidism. The only example of a strong environmental influence was found in extreme maternal and fetal iodine deficiency that led to cretinism; but this is of no concern in countries with iodine supplementation and newborn-screening programs. However, in contrast to manifest hypothyroidism, slight elevations of TSH have been monitored in areas with milder iodine deficiency. In the paper by Baccarelli et al., a new cause of neonatal TSH change – however in the normal range! – was described in a population of neonates from mothers with dioxin exhibition in the Seveso nuclear accident 30 years ago. Due to the extremely long half-life of digoxin, the authors could still measure relevant levels of the toxin in the maternal blood and it seems that the fetal thyroid axis is mildly influenced by the toxin; the neonatal TSH correlated with the level of maternal toxin load. While it remains to be determined how digoxin affects thyroid function, the good news is that even in highly exposed women, children are *not* born with congenital hypothyroidism and so far there is no evidence to argue that a mildly elevated TSH within the normal range may cause harm to the developing child.

This discussion of cognitive outcome and mild neonatal differences in thyroid function has seen a significant improvement by the following study of a large cohort of children tested for cognitive function in relation to neonatal T_4 values.

Neonatal thyroxine, maternal thyroid function, and child cognition

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J Clin Endocrinol Metab 2009;94:497–503

Background: Thyroid hormone is essential for normal brain development. Limited data are available regarding whether thyroid function in neonates influences later cognitive development.

Methods: The authors studied cognitive test scores at ages 6 months and 3 years in participants in Project Viva, a cohort study in Massachusetts with a total of 500 children born 1999–2003 at 34 weeks or more.

Results: Mean newborn T_4 at a mean age of 1.94 days was 17.6 (SD 4.0) $\mu\text{g/dl}$, and levels were higher in girls [1.07 $\mu\text{g/dl}$; 95% CI 0.38, 1.76] and infants born after longer gestation (0.42 $\mu\text{g/dl}$; 95% CI 0.17, 0.67 per week). Newborn T_4 levels were not associated with maternal T_4 , TSH, or thyroid peroxidase antibody levels. On multivariable linear regression analysis, adjusting for maternal and child characteristics, higher newborn T_4 was unexpectedly associated with poorer scores on the visual recognition memory test among infants at age 6 months (-0.5 ; 95% CI -0.9 , -0.2), but not with scores at age 3 years on either the Peabody Picture Vocabulary Test (0.2; 95% CI -0.1 , 0.5) or the Wide Range Assessment of Visual Motor Abilities (0.1; 95% CI -0.2 , 0.3). Maternal thyroid function test results were not associated with child cognitive test scores.

Conclusions: Newborn T_4 concentrations within a normal physiological reference range are not associated with maternal thyroid function and do not predict cognitive outcome in a population living in an iodine-sufficient area.

The very important and clear message of this study is that irrespective of the TSH level – unfortunately we do not even know the TSH levels of the investigated children – the cognitive development of neonates is *not* related to the concentration of neonatal T_4 as long as it is within the normal range. It seems to make no difference if a child was born with a high normal or low normal T_4 and this correlates to a wide range from 6.4 to 35.7 $\mu\text{g/dl}$. Moreover, the neonatal T_4 value and cognitive development of the newborns did not correlate with maternal thyroid function in the first trimester. Obviously these data do not completely exclude those children with mild thyroid dysfunction in terms of isolated TSH elevation and normal T_4 ('hyperthyrotropinemia') who will all have a normal cognitive outcome. But concerns about children with mild TSH elevation, as long as T_4 is normal, need to be downgraded. Moreover, the trend to lower TSH cut-off levels in neonatal screening programs [9] needs to be reconsidered in regard to the normal outcome of these 500 studied newborns. Beside the very sensitive question of hyperthyrotropinemia in newborns, elevated TSH in older children is a very frequent cause to seek a pediatric endocrinologist, and data are needed to have good ground for clinical decisions what to do with these children of mostly incidental findings of elevated TSH. Two studies were published in the last 12 months that contribute significantly to this important clinical question.

Prospective evaluation of the natural course of idiopathic subclinical hypothyroidism in childhood and adolescence

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Eur J Endocrinol 2009;160:417–421

Background: The authors aimed to prospectively evaluate the course of subclinical hypothyroidism (SH) in children and adolescents with no underlying diseases and no risk factors, which might interfere with the progression of SH.

Methods: Clinical status, thyroid function, and autoimmunity were prospectively evaluated at entry and after 6, 12, and 24 months in 92 young patients (mean age 8.1 ± 3.0 years) with idiopathic SH.

Results: During the study, mean TSH levels showed a trend toward a progressive decrease while FT_4 levels remained unchanged. Overall, 38 patients normalized their TSH (group A): 16 patients between 6 and 12 months, and 22 patients between 12 and 24 months. Among the remaining 54 patients, the majority maintained TSH within the baseline values (group B), whereas 11 exhibited a further increase in TSH >10 mU/l (group C). Baseline TSH and FT_4 levels were similar in the patients who normalized TSH, compared with those with persistent hyperthyrotropinemia. Even in the patients of group C, both TSH and FT_4 at entry were not different with respect to those of groups A and B. No patients showed any symptoms of hypothyroidism during follow-up and no changes in both height and body mass index were observed throughout the observation period.

Conclusion: The natural course of TSH values in a pediatric population with idiopathic subclinical hypothyroidism is characterized by a progressive decrease over time; the majority of patients (88%) normalized or maintained unchanged their TSH, and TSH changes were not associated with either FT_4 values or clinical status or auxological parameters.

Natural history of thyroid function tests over 5 years in a large pediatric cohort

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J Clin Endocrinol Metab 2009;94:1678–1682

Background: Because clinical manifestations of thyroid disorders are variable and subtle in children and adolescents, thyroid function tests are often repeated in patients with nonspecific symptoms. The objective of the study was to determine the natural history of initial abnormal TSH and define populations at greater risk for developing a subsequent thyroid dysfunction.

Methods: A total of 121,052 of 1.043 million outpatients aged 0.5–16 years insured by the Clalit Health Medical Organization had a TSH determination in 2002 and follow-up to 2007. Extracted from the

Clalit Health Medical Organization database were their demographic data, referral diagnoses, and laboratory results (TSH, free T_4 , thyroid antibodies). Excluded were patients with overt hypothyroidism or hyperthyroidism on initial testing.

Results: Results of 96.5% of initial serum TSH concentrations were normal (0.35–5.5 mIU/l), 0.2% were low (<0.35 mIU/l), 2.9% elevated (>5.5 to ≤10 mIU/l), and 0.4% highly elevated (>10 mIU/l). The frequency of TSH testing increased with age and female gender. During follow-up, repeated (two to more than four) TSH tests were performed in 45.7% of the patients. In the second TSH determination, normal TSH was documented in 40, 73.6, and 78.9% of those whose initial serum TSH was highly elevated, elevated, and low, respectively, and in 97% of those with normal initial TSH. Predictive factors for a sustained highly elevated TSH were initially TSH >7.5 mIU/liter ($p = 0.014$) and female gender ($p = 0.047$).

Conclusion: In the pediatric population, initial normal or slightly elevated TSH levels are likely to remain normal or spontaneously normalize without treatment. Patients with initial levels >7.5 mIU/l, particularly girls, are at a greater risk for sustained abnormal TSH levels.

Both studies give evidence that an elevated TSH in the range of 5–10 and higher while T_4 is normal seems not to be a cause for concern in children. Based on a prospective follow-up study of 92 patients – including measurement of antibodies and thyroid ultrasound – from Italy as well as a huge study from Israel based on 121,052 (!) children, the authors all together demonstrated that almost all children did not develop overt thyroid disease within the study period of 0.5–16 years and that 50–70% of elevated TSH normalized just by ‘observation’. Those children who maintained with an elevated TSH mostly did not further increase and only 2% developed autoimmune thyroiditis in the Italian study and 0.4% developed manifest hypothyroidism in the study from Israel.

As a conclusion, we have again to accept that Nature is not exact. TSH levels vary and the arithmetic border of normal, which defines a TSH >2 standard deviations as ‘unnormal’, does *not* define a disease for itself. We should be conservative in our management of elevated TSH while T_4 is normal.

Extreme longevity is associated with increased serum thyrotropin

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J Clin Endocrinol Metab 2009;94:1251–1254

Background: The distribution of serum TSH shifts progressively to higher concentrations with age. The aim of the study was to determine whether the population shift in TSH distribution to higher concentrations with aging extends to people of exceptional longevity, namely centenarians, and to assess the relationship between concentrations of TSH and free T_4 (FT_4).

Methods: The authors analyzed TSH, FT_4 , and TSH frequency distribution curves in thyroid disease-free Ashkenazi Jews with exceptional longevity (centenarians; median age 98 years), in younger Ashkenazi controls (median age 72 years), and in a population of thyroid disease-free individuals (median age 68 years) from the US National Health and Nutrition Examination Survey 1998–2002 (NHANES controls).

Results: Serum TSH was significantly higher in centenarians [1.97 (0.42–7.15) mIU/l] than in Ashkenazi controls [1.55 (0.46–4.55) mIU/l] and NHANES controls [1.61 (0.39–6.29) mIU/l] (median 2.5 and 97.5 centiles) ($p < 0.001$). The TSH frequency distribution curve of centenarians was relatively similar in shape to controls but shifted significantly to higher TSH, including TSH concentration at peak frequency. The TSH distribution curve of the NHANES control group was superimposable to and not significantly different from the Ashkenazi controls. FT_4 was similar in centenarians and Ashkenazi controls, and there was a significant inverse correlation between FT_4 and TSH in both groups.

Conclusions: The TSH population shifts to higher concentrations with age appear to be a continuum that extends even to people with exceptional longevity. The inverse correlation between TSH and FT_4 in these populations suggests that changes in negative feedback may contribute to exceptional longevity.

Although pediatric endocrinologists are not frequently confronted with patients older than 100 years of age, it might be interesting for our community to know that higher TSH levels do not seem to be a cause for concern when thinking of a long life expectancy. The lucky individuals who reach

such exceptionally old age seem to have higher TSH levels. However we do not know if higher TSH values are essential for reaching such an old age, but at least they do not seem to detain a long life expectancy.

New hope: to predict the outcome of Graves' disease in children

Predictors of autoimmune hyperthyroidism relapse in children after discontinuation of antithyroid drug treatment

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Background: There is debate about how Graves' disease (GD) should be treated in children. The aim of this study was to identify predictors of relapse after antithyroid drug (ATD) treatment in children with GD.

Methods: The authors conducted a prospective, multicenter cohort study of children ($n = 154$) with GD treated with carbimazole for an intended duration of 24 ± 3 months. After the end of treatment, patients were followed up for at least 2 years. The primary outcome was hyperthyroidism relapse. Cox's regression analysis was used and a prognostic score was constructed.

Results: The overall estimated relapse rate for hyperthyroidism was 59% (95% CI 52–67%) at 1 year and 68% (95% CI 60–76%) at 2 years after the end of treatment. Multivariate survival analysis showed that the risk of relapse was higher for patients of non-Caucasian origin [hazard ratio (HR) = 2.54, $p < 0.001$], with high serum thyroid-stimulating hormone receptor antibodies (HR = 1.21 by 10 U, $p = 0.03$) and free T_4 (HR = 1.18 by 10 pmol/l, $p = 0.001$) levels at diagnosis. Conversely, relapse risk decreased with increasing age at onset (HR = 0.74 per 5 years, $p = 0.03$) and duration of first course of ATD (HR = 0.57 per 12 months, $p = 0.005$). A prognostic score was constructed, allowing the identification of three different risk groups, with 2-year relapse rates of 46, 77, and 98%.

Conclusion: A longer initial duration of euthyroid state with ATD seems to be the only variable related to the risk of hyperthyroidism relapse in children that can be manipulated. Ethnic origin, age, and severity of the disease at diagnosis may guide long-term disease management decisions.

Since the initial and provocative publication by Barbara Lippe in 1987 which claimed a close correlation of initial duration of antithyroid drug treatment and long-term outcome of Grave's disease, no final proof for that hypothesis was contributed to the field. Although not closing the gap of evidence, the French release of follow-up data from 154 children with Grave's disease – which represents the largest pediatric cohort published ever – is very good ground to prolong medication before deciding for definite treatment by surgery or radioiodine. The retrospective data show that medical treatment for over 24 months can reduce the 'relapse prediction score' from 98 to 44%. This needs to be confirmed in a prospective intervention study comparing 24 versus 24+ months of treatment. However, this study has been due since Lippe's data in 1987 – let's do it now!

Whoever will do this study and whoever treats children with antithyroid drugs, it should be methimazole and not propylthiouracil. This statement is based on the 'papers' that appeared as a report of a meeting rather than a meta-analysis study in the *JCEM* and in the *NEJM*. The authors reported data that were collected from different US institutions showing that liver failure as a severe adverse event of antithyroid drug treatment only occurred when PTU was given. Although the number of liver failures was very low with an incidence of 1 in 2,000–4,000 treated cases, all cases were under PTU and it seems that this side effect can be circumvented by prescribing methimazole. Regardless of the fact that no official study has been conducted, the two releases have such a clinical impact that they are cited here in the Yearbook.

Putting propylthiouracil in perspective

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J Clin Endocrinol Metab 2009;94:1881–1882

Ending propylthiouracil-induced liver failure in children

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N Engl J Med 2009;360:1574–1575

References

1. De Felice M, Di Lauro R: Thyroid development and its disorders: genetics and molecular mechanisms. *Endocr Rev* 2004;25:722–746.
2. Rallison ML, Dobyns BM, Keating FR Jr, et al: Thyroid nodularity in children. *JAMA* 1975;233:1069–1072.
3. Reiners C, Wegscheider K, Schicha H, et al: Prevalence of thyroid disorders in the working population of Germany: ultrasonography screening in 96,278 unselected employees. *Thyroid* 2004;14:926–932.
4. Yang J, Schnadig V, Logrono R, Wasserman PG: Fine-needle aspiration of thyroid nodules: a study of 4,703 patients with histologic and clinical correlations. *Cancer* 2007;111:306–315.
5. Eden K, Mahon S, Helfand M: Screening high-risk populations for thyroid cancer. *Med Pediatr Oncol* 2001;36:583–591.
6. Overbeck B, Sundet K, Kase B, Heyerdahl S: Congenital hypothyroidism: Influence of disease severity and L-thyroxine treatment on intellectual, motor and school-associated outcomes in young adults. *Pediatrics* 2003;112:923–930.
7. Rovet JF: Children with congenital hypothyroidism and their siblings: Do they really differ? *Pediatrics* 2005;115:e52–e57.
8. Selva KA, Harper A, Downs A, Blasco PA, Lafranchi SH: Neurodevelopmental outcomes in congenital hypothyroidism: comparison of initial T₄ dose and time to reach target T₄ and TSH. *J Pediatr* 2005;147:775–780.
9. Corbetta C, Weber G, Cortinovis F, Calebiro D, Passoni A, Vigone MC, Beck-Peccoz P, Chiumello G, Persani L: A 7-year experience with low blood TSH cutoff levels for neonatal screening reveals an unsuspected frequency of congenital hypothyroidism. *Clin Endocrinol (Oxf)* 2009, Epub ahead of print.