Available online at www.sciencedirect.com

ScienceDirect

Journal of the Chinese Medical Association 78 (2015) 145–153

www.jcma-online.com

Review Article

A systematic review of genetic studies of thyroid disorders in Taiwan

Chun-Jui Huang [a,b], Tjin-Shing Jap [a,b,*]

[a] *Division of Endocrinology and Metabolism, Department of Medicine, Taipei Veterans General Hospital, Taipei, Taiwan, ROC*
[b] *Faculty of Medicine, National Yang-Ming University, Taipei, Taiwan, ROC*

Received January 17, 2014; accepted August 5, 2014

Abstract

A systematic review of genetic studies of thyroid disorders in Taiwan identified studies of gene mutations involved in the synthesis and binding of thyroid hormone, as well as mutations of proto-oncogenes and tumor suppressor genes in thyroid cancer. Studies related to gene polymorphisms in patients with autoimmune thyroid disease (AITD) and thyroid cancer were also reviewed. The most prevalent mutations in the Han-Chinese population were c.2268insT in the thyroid peroxidase (*TPO*) gene and c.919-2A>G in the Pendred syndrome (*PDS*) gene. Additional mutations have also been revealed in the genes encoding TPO ($n = 5$), thyroglobulin (TG; $n = 6$), pendrin ($n = 2$), and thyroxine-binding globulin (TBG; $n = 2$), which were novel at the time they were reported. The prevalence of various somatic mutations in differentiated thyroid cancer was similar in Taiwan and Western countries, with the *RAS kinase* mutation and tyrosine receptor kinase (*TRK*) and rearranged during transfection (*RET*) proto-oncogenes being detected in lower frequencies and the B-type RAF kinase (*BRAF*) mutation accounting for the majority of cases. Recent microRNA analysis revealed an association between miR146b and the *BRAF* mutation, which was associated with poor prognosis of papillary thyroid carcinoma (PTC). Susceptibility to Graves' disease (GD) was linked to the human leukocyte antigen (HLA) region. The associated alleles were different in Han-Chinese and Caucasians; HLA-DPB1*0501, the major allele in Taiwan, has a low frequency in the West. By contrast, a high frequency of HLA-DRB1*0301 was detected in Caucasians but not Han-Chinese. In addition to the HLA region, cytotoxic T lymphocyte-associated molecule-4 (*CTLA4*) gene polymorphisms +49G>A and +6230G>A (CT60) were positively associated with GD. The GG genotype and G allele of single nucleotide polymorphism (SNP) +49G>A were also related to relapse of Graves' hyperthyroidism after antithyroid drug withdrawal. Differences in the genetic patterns between Han-Chinese and Caucasians for some thyroid disorders suggest the importance of variable genetic influences in different populations.

Keywords: gene; mutation; Taiwan; thyroid

1. Introduction

Thyroid disorders may be categorized into two general groups, including functional disorders such as hyperthyroidism and hypothyroidism, and structural abnormalities, including goiter and thyroid neoplasia. The basis of some thyroid disorders may reflect the effect of a mutation in a single gene (e.g., monogenic) or the effects of polymorphisms in multiple genes. Among the monogenic diseases, germline mutations can affect thyroid hormone synthesis or thyroid hormone binding in serum. However, somatic mutations play a major role in thyroid neoplasia pathogenesis. Impaired synthesis of thyroid hormone may result in congenital hypothyroidism while mutations in the genes encoding thyroid hormone carrier proteins lead to clinically euthyroid patients with falsely abnormal thyroid function tests. In addition, an increased risk for autoimmune thyroid disease (AITD) or thyroid neoplasia has been associated with genetic polymorphisms at various loci (Fig. 1). The current review will

Conflicts of interest: The authors declare that there are no conflicts of interest related to the subject matter or materials discussed in this article.

* Corresponding author. Dr. Tjin-Shing Jap, Division of Endocrinology and Metabolism, Department of Medicine, Taipei Veterans General Hospital, 201, Section 2, Shih-Pai Road, Taipei 112, Taiwan, ROC.
E-mail address: tsjap@vghtpe.gov.tw (T.-S. Jap).

http://dx.doi.org/10.1016/j.jcma.2014.09.010

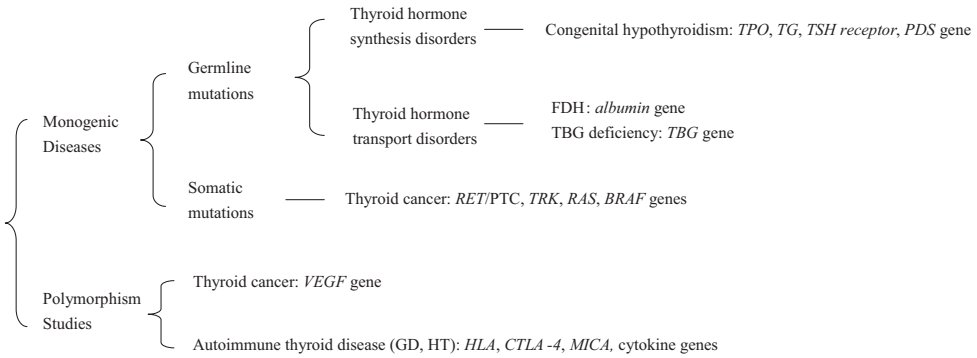

Fig. 1. Schematic diagram for genetic studies of thyroid disorders in Taiwan. BRAF = B-type RAF kinase; CTLA-4 = cytotoxic T lymphocyte-associated molecule-4; FDH = familial dysalbuminemic hyperthyroxinemia; GD = Graves' disease; HLA = human leukocyte antigen; HT = Hashimoto thyroiditis; MICA = major histocompatibility complex class I-chain related gene A; PDS = pendred syndrome/pendrin; PTC = papillary thyroid carcinoma; RET = rearranged during transfection; TBG = thyroxine-binding globulin; TG = thyroglobulin; TPO = thyroid peroxidase; TRK = tyrosine receptor kinase; TSH = thyroid stimulating hormone; VEGF = vascular endothelial growth factor.

summarize the results of major genetic studies performed in Taiwan regarding thyroid disorders to provide a basis for further understanding of the thyroid genetics in the Han-Chinese population.

2. Genetic basis of congenital hypothyroidism

The prevalence of congenital hypothyroidism in Taiwan ranges from 1 in 1992 live births to 1 in 5788 live births,[1,2] which is similar to the 1 in 4000 live births reported in European and North American populations.[3,4] Thyroid dysgenesis, such as agenesis, hypoplasia, ectopy, and hemiagenesis, accounts for 80−85% of congenital hypothyroidism; thyroid dyshormonogenesis contributes to the remaining 15−20% of cases.[5,6] Congenital hypothyroidism may be the result of impaired thyroid stimulating hormone (TSH) signaling, abnormal thyroid hormone synthesis, or defective thyroid hormone action in target tissues (Fig. 2).[6] The majority of genetic studies in Taiwan have focused on germline mutations involving the thyroid hormone synthesis pathways, such as pendrin for the trapping of iodide into thyroid follicular cells, and thyroid peroxidase (TPO), which regulates iodide organification by binding iodide to thyroglobulin (TG) and further coupling to form thyroxine (T4) and triiodothyronine (T3).[7−9]

The severity of thyroid dyshormogenesis is based on whether there is a total or partial defect in the organification of iodide. Mutation in the *TPO* gene is the major contributing factor for total iodide organification defect (TIOD); mutation in the Pendred syndrome (*PDS*) gene usually causes partial iodide organification defect (PIOD).[7,10] Despite an intact thyroid hormone synthesis pathway in the thyroid follicular cells, synthesis of thyroid hormone remains impaired in the absence of a signal from pituitary thyroid stimulating hormone (TSH) to a functional thyroid TSH receptor. Loss-of-function mutations in the *TSH receptor* gene may induce TSH resistance and subsequent congenital hypothyroidism.[11] Mutations in the thyroid hormone receptor alpha and beta have been reported to result in impaired thyroid hormone action in target tissues; however, we failed to identify any study in Taiwan regarding this issue.[12,13]

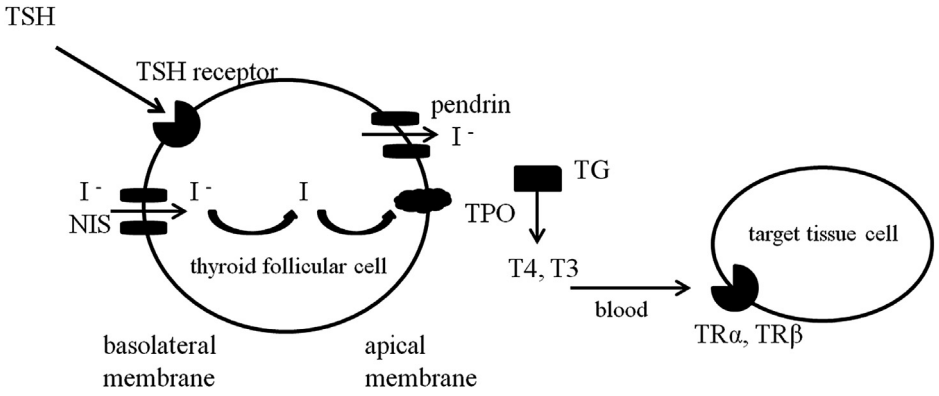

Fig. 2. Mutations that may lead to congenital hypothyroidism include those in the TSH receptor causing impaired TSH signaling, mutations involved in the synthesis of thyroid hormone, and mutations in TRα and TRβ, causing defective thyroid hormone action in target tissues. Key elements involved in thyroid hormone synthesis include the NIS and pendrin for iodide trapping, TPO for facilitating iodide binding to TG, and further coupling to form T4 and T3. NIS = sodium iodide symporter; TG = thyroglobulin; TPO = thyroid peroxidase; TRα = thyroid hormone receptor alpha; TRβ = thyroid hormone receptor beta; TSH = thyroid stimulating hormone; T3 = triiodothyronine; T4 = thyroxine.

2.1. TPO

The human *TPO* gene consists of 17 exons and spans about 150 kilobases on chromosome 2p25.[14,15] In the study by Niu et al[7] that evaluated 16 patients from 16 unrelated Taiwanese families with thyroid dyshormogenesis, seven patients were classified as having TIOD, and two patients had PIOD as detected by the perchlorate discharge test. Three mutations, including c.2268insT, c.2243delT, and p.G157C, in the *TPO* gene were identified in the seven patients with TIOD; no *TPO* gene mutation was detected in the two patients with PIOD.[7] In another study from Taiwan, five *TPO* gene mutations, including c.2268insT, c.843delC, c.2413delC, c.1477G>A, and c.2386G>T, were detected in five unrelated patients with TIOD.[8] All of the above mutations identified in Taiwanese patients had not been detected in other ethnic groups at the time of the report, suggesting the heterogenic nature of *TPO* mutations by ethnicity. The c.2268insT mutation was found to be the most common mutation observed in both studies, accounting for 86% (12/14) of the mutant alleles in the report from Niu et al[7] and 40% (4/10) of the studied alleles in Wu et al.[8] A thymidine (T) insertion in exon 13 of the 2268 residue of the *TPO* gene may lead to a premature stop codon, resulting in the formation of truncated polypeptides and dysfunctional TPO enzymes.[7,8] In addition to being the cause of congenital hypothyroidism, c.2268insT was also associated with transient hypothyroidism of the neonate.[16] Haplotype analysis revealed that the c.2268insT mutation in Taiwan may be due to a founder effect.[7]

2.2. PDS

The *PDS* gene, also known as *SLC26A4*, contains 21 exons expanding 57,174 base pairs on chromosome 7q22-q31.1.[17–19] This gene encodes the pendrin protein, an anion transporter expressed in the thyroid, kidney, and inner ear. Pendred syndrome (PS) is an autosomal recessive disorder caused by biallelic mutations in the *PDS* gene, manifesting as congenital sensorineural deafness, goiter, partial iodine organification defect, and thyroid dysfunctions, ranging from euthyroid to hypothyroidism.[10] High heterogeneity and differences by ethnicity were noted in the PDS mutations. Huang et al[20] recently summarized the mutation spectrum of the *SLC26A4* gene in the Chinese population. Although the great majority of mutations found worldwide were missense mutations, a single splice-site mutation of c.919-2A>G (g.IVS7-2A>G) was reported to account for > 80% of the mutations observed in Taiwan.[21–23] Following c.919-2A>G, c.2168A>G (p. H723R) was the second most common mutation in Han-Chinese.[20] By contrast, the p.L236P, p.T416P, IVS8+1G>A, p.E384G, p.L445W, p.T410M, p.G209V, p.V138F, p.Y530H, and p.L597S mutations were more commonly detected in Caucasians.[20] In another study from Taiwan by Lai et al,[23] a missense mutation of c.1079C>T (p.A360V) in exon 9 of the *SLC26A4* gene was discovered for the first time in 2007. Huang et al[20] later identified another new splice-site mutation of c.1263+1G>A (g.IVS10+1G>A) in compound heterozygosity with c.1079C>T in a Taiwanese patient with PS.

2.3. TG

Human TG is encoded by a large gene of 270 kilobases that maps to chromosome 8q24 and contains a coding sequence of 8.5 kilobases divided into 48 exons.[24] Mutations in the *TG* gene may result in congenital hypothyroidism, goiter, thyroid neoplasia, or familial AITD.[25] In seven Taiwanese patients from six families with a TG defect, six new *TG* gene mutations, including c.1348delT, p.R432X (c.1351C>T), g.IVS3+2T>G, c.1712delT, p.Q1765X (c.5350C>T), and c.6047delA, were identified, which all led to a premature translation termination.[9] The most common mutations were c.1348delT and p.R432X.[9] The high prevalence of c.1348delT was thought to be due to a founder effect by linking to a specific haplotype, and p.R432X was due to an independently recurrent *de novo* mutation, suggesting a mutational hot spot.[9]

2.4. TSH receptor mutation

The human *TSH receptor* gene is located on chromosome 14q31 and encodes the G-protein-coupled TSH receptor from exons one to 10.[9] Autonomous thyroid adenoma or congenital hyperthyroidism may develop as a result of gain-of-function mutations, and loss-of-function mutations in the *TSH receptor* gene are the most common cause of TSH resistance.[26] Systematic analysis of the *TSH receptor* gene in Japanese patients with congenital hypothyroidism revealed a particularly prevalent variant c.1349G>A (p.R450H) as the cause of loss-of-function *TSH receptor* mutations.[27] This mutation was also shown to be important in Taiwanese children with congenital hypothyroidism.[28]

3. Genetic alterations affecting thyroid hormone binding

The physiologically active forms of the thyroid hormones, free T4 and free T3, account for around 0.3% of the circulating thyroid hormones; the remaining 99.7% are bound to carrier proteins, including thyroxine-binding globulin (TBG; 70%), albumin (15–20%), and transthyretin (10–15%). Patients with carrier protein gene mutations are clinically euthyroid but laboratory abnormal and are often misdiagnosed as having thyroid dysfunction.[29,30] In patients with familial dysalbuminemic hyperthyroxinemia (FDH), serum albumin exhibits increased binding affinity to T4, causing free T4 and total T4 concentrations to be elevated as detected by the one-step analog method but normal using the equilibrium dialysis or two-step methods.[31,32] In TBG deficiency, a low concentration of total T4 and total T3 may result in the misdiagnosis of hypothyroidism if free T4 was not measured simultaneously or subsequently, resulting in unwarranted treatment with antithyroid drug or thyroid hormone replacement.[30]

3.1. FDH

The human *albumin* gene is located within q11-22 of chromosome 4 and contains 15 exons split by 14 intervening

sequences.[33] The molecular basis of FDH relates to a point mutation in codon 218 of the *albumin* gene, which causes a substitution of histidine (CAC) or proline (CCC) for arginine (CGC).[34,35] The first Chinese case of FDH with guanine (G) to adenosine (A) transition in the second nucleotide of codon 218 of the *albumin* gene was reported in 1999.[29] This missense mutation is the same as that described frequently in Western white families, but differs from the one found in a Japanese kindred analysis.[34,35]

3.2. TBG deficiency

TBG is encoded by the *TBG* gene on Xq22 chromosome and consists of five exons spanning 5.5 kbp.[36,37] TBG anomalies are transmitted through an X-linked fashion and produce the following three clinical phenotypes determined by the serum levels of TBG: complete TBG deficiency, partial TBG deficiency, and TBG excess.[38,39] Complete TBG deficiency is caused by either early termination of translation or an amino acid substitution, resulting in secretion failure.[40,41] In the report by Su et al,[30] two *TBG* gene mutations, which were novel at that time, were discovered in two cases of complete TBG deficiency in Taiwan. In one case, a missense mutation, p.S52N, consisting of a G to A transition in codon 52, resulted in the substitution of serine (AGC) for asparagine (AAC). In the other case, a nonsense mutation, p.W280X, consisting of a G to A transition in codon 280 (TGG→TGA) resulted in premature termination of translation and therefore, a truncated protein.[30]

4. Somatic mutations in thyroid cancer

Thyroid cancers arising from thyroid epithelial cells include the differentiated thyroid cancers papillary thyroid carcinoma (PTC), follicular carcinoma, and poorly differentiated carcinoma, and the undifferentiated thyroid cancer anaplastic thyroid carcinoma (ATC). PTC and follicular carcinoma account for > 90% of thyroid cancers and generally carry a favorable prognosis with 10-year survival rates of 93% and 85%, respectively.[42] By contrast, patients with ATC have a median survival of < 6–8 months.[43]

Our current understanding of the molecular pathogenesis of thyroid cancer resulted from identification of genetic and epigenetic alterations in various signaling pathways, including the mitogen activated protein kinase (MAPK), the phosphatidylinositol-3 kinase (PI3K)–protein kinase (AKT), the nuclear factor-κB (NF-κB), the Ras association domain-containing protein 1 (RASSF1)-mammalian sterile 20 (STE20)-like protein kinase 1 (MST1)–forkhead box O3 (FOXO3), the WNT-β-catenin, the hypoxia-inducible factor 1α (HIFIα), and the TSH receptor pathways.[44] Among the various pathways, the MAPK and PI3K–AKT are the pathways best characterized and show promise in thyroid cancer treatment.[45] Activation of the MAPK pathway mainly drives the development of PTC.[44] By contrast, activation of the PI3K–AKT pathway is involved in the formation of follicular adenoma and carcinoma.[44] Upon simultaneous activation of both pathways with an accumulation of genetic alterations, PTC or follicular carcinoma may progress to poorly differentiated carcinoma or anaplastic carcinoma.[44]

4.1. Mutations involving the MAPK–PI3K–AKT pathway

MAPK–PI3K–AKT signaling is essential for maintaining cell growth, proliferation, differentiation, and apoptosis (Fig. 3).[46] Approximately 70% of cases of PTC are related to somatic mutations involving the RET/PTC→RAS→-BRAF→mitogen extracellular kinase (MEK)→extracellular signal-regulated kinase (ERK) pathway.[44,47] Rearrangement of two receptor tyrosine kinase genes, rearranged during transfection (*RET*) and neurotrophic tyrosine kinase receptor type 1 (*NTRK1*), are specifically expressed in PTC.[48,49] The frequency of *RET*/PTC rearrangement ranges from 55% ($n = 6/11$) as reported by Lee et al[50] to 8% ($n = 8/105$) in Liu et al.[51] After comparing its prevalence, which ranges from 0% to 20% worldwide,[52–54] we think that the *RET*/PTC may not

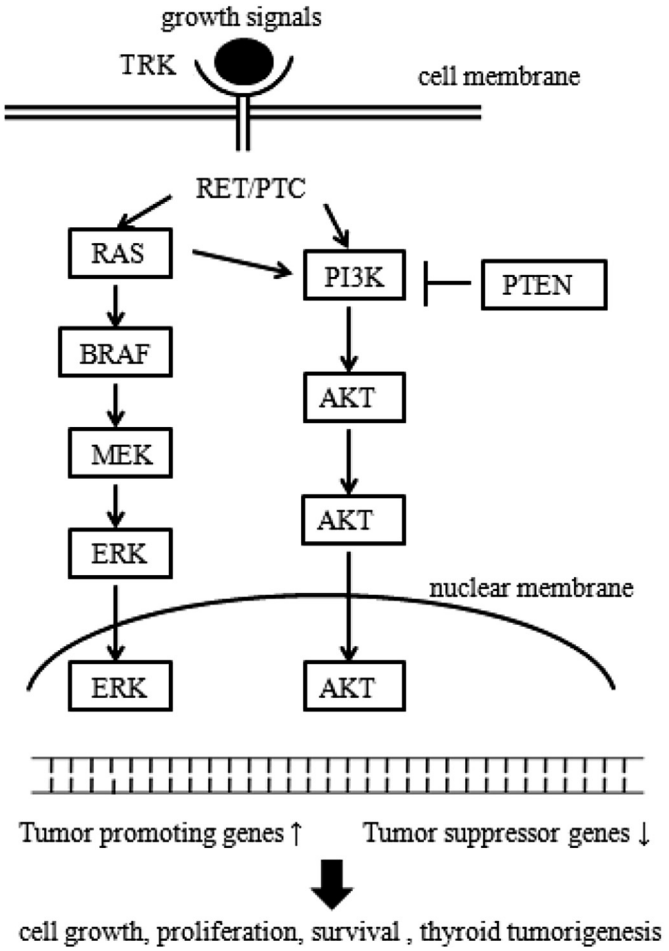

Fig. 3. The MAPK–PI3K–AKT pathway. AKT = protein kinase; BRAF = B-type RAF kinase; ERK = extracellular signal-regulated kinase; MAPK = mitogen activated protein kinase; MEK = mitogen extracellular kinase; PI3K = phosphatidylinositol-3 kinase; PTC = papillary thyroid carcinoma; PTEN = phosphatase and tensin homolog; RET = rearranged during transfection; TRK = tyrosine receptor kinase.

be as common as previously reported by Lee et al.[50] Moreover, the chimeric gene resulting from *NTRK1* rearrangement, the *TRK* proto-oncogene, was not detected in 40 thyroid tumors isolated from patients in Taiwan, suggesting that the prevalence of *TRK* mutation is even lower than *RET*/PTC rearrangement in Taiwan.[51,55] In a study of 89 thyroid tumors, four *RAS* mutations were detected in follicular carcinoma ($n = 3$) and follicular adenoma ($n = 1$); however, they were absent in the papillary carcinomas analyzed, suggesting that its occurrence differs by tumor type.[56]

In contrast to the low occurrence of *RET*/PTC rearrangement and the *TRK* and *RAS* mutations in Taiwan, the *BRAF* mutation was reported to be the most prevalent mutation in PTCs and found exclusively in thyroid cancers arising from papillary cells.[47,57,58] The heterozygous mutation c.T1799A (p.V600E) in exon 15 of the *BRAF* gene was detected in 49 of 105 (47%) sporadic cases of PTC in Taiwan,[57] which was a similar frequency to that reported from other geographical areas.[59–61] Recent meta-analysis suggested a positive correlation between *BRAF* mutation and high-risk clinicopathological features of PTC, such as extrathyroid invasion, lymph node metastasis, and advanced tumor stage.[62] However, inconsistency exists in the literature,[47] and no correlation was found in a study conducted in Taiwan.[57] Recent studies on microRNA deregulation have demonstrated an association of specific microRNAs, particularly miR-146b, with the aggressiveness and prognosis of PTC,[63,64] which may be related to increased cell proliferation, migration, invasion, or resistance to chemotherapy-induced apoptosis in response to aberrant microRNA expression.[64]

4.2. Mutations outside the MAPK pathway

In addition to mutations related to the MAPK pathway, other genetic alterations have been postulated to have a role in the pathogenesis of thyroid cancer, including those affecting the *p53*, phosphatase and tensin homolog (*PTEN*), and fragile histidine triad protein (*FHIT*) genes.[65–67] It is understood that p53 suppresses growth in normal cells; its inactivation is associated with colon, lung, breast, and thyroid cancer development.[68] In Taiwan, mutations in the *p53* gene have been detected in five of 29 (17.2%) cases of poorly differentiated carcinomas, two of 41 (5.0%) cases of well-differentiated papillary carcinoma, and one of six (16.7%) cases of oncocytic carcinoma of the thyroid.[65] Consistent with the results of other reports, the *p53* gene mutations identified included single nucleotide changes, resulting in missense or nonsense mutations.[65]

Germline mutations in the *PTEN* tumor suppressor gene occur often in Cowden syndrome, a syndrome characterized by multiple tumor-like harmatomas and an increased risk of certain cancers, such as breast, endometrial, and thyroid follicular carcinomas.[69] However, somatic mutation of the *PTEN* gene was not found in any of the 17 sporadic thyroid tumors studied in Taiwan, indicating that the *PTEN* gene may not play a major role in sporadic thyroid tumors.[66] With complete sequencing of the *FHIT* gene, deletions between exons 2 and 9 were found in seven cases of thyroid tumors in Taiwan, suggesting that *FHIT* gene alterations may have a role in the formation of thyroid neoplasm.[67]

4.3. Mutation analysis in anaplastic thyroid carcinoma

Mutation analysis of 16 anaplastic thyroid carcinoma patients in Taiwan revealed five (5/16, 31%) cases with an *N-RAS* mutation and one (1/16, 6%) case with a *BRAF* mutation.[70] While the current understanding is that accumulation mutations in both the MAPK and PI3K–AKT pathways may result in anaplastic thyroid carcinoma, this disease may arise more often from *RAS*-mutant tumors than from *BRAF*-mutant tumors.[70]

4.4. Genetics of medullary thyroid carcinoma

Medullary thyroid carcinomas (MTCs) derive from parafollicular calcitonin-producing C-cells originating from the neural crest.[71] MTC is a type of neuroendocrine tumor that possesses distinct clinical and pathological characteristics from differentiated thyroid cancers.[71] The genetic basis of MTC is different from that of the differentiated thyroid cancers due to its association with multiple endocrine neoplasia (MEN) and familial MTC syndrome.[72] Mutations in the *RET* proto-oncogene were detected in > 95% of patients with MTC with MEN type 2 (MEN-2), but the frequency dropped to around 50% in sporadic MTCs.[72] Consistent with the results reported for Western countries, mutation analysis of the *RET* proto-oncogene in sporadic and MEN-associated MTC in Taiwan revealed two common mutation sites at codon 634, causing a substitution of cysteine to arginine or phenylalanine (p.C634R, p.C634Y), and at codon 918, resulting in methionine to threonine replacement (p.M918T).[73–75] Furthermore, in cytomorphology analysis of MTCs using fine needle aspiration specimens, codon 634 mutations were more strongly associated with small/round or large/oval to polygonal cells, while codon 918 mutations were more related to small/round and spindle shape cells.[75]

5. Polymorphism studies in thyroid cancer

In Taiwan, gene polymorphisms of the vascular endothelial growth factor (*VEGF*) gene, genes of the BER (base excision repair) pathway, and the glutathione peroxidase 3 (*GPX3*) gene have been studied in thyroid cancer.[76–79] VEGF acts through the transmembrane fms-like tyrosine kinase (FLT) receptor family to influence cell proliferation, migration, and angiogenesis; it also plays an important role in thyroid cancer cell growth and distant metastasis.[76] Analysis of functional single nucleotide polymorphisms (SNPs) of *VEGF* in Taiwan disclosed that the A allele of K2578C/A (i.e., SNP rs699947) was associated with an increased risk of thyroid cancer and regional lymph node metastasis in men.[77] Alterations in genes associated with the BER pathway, one of the DNA repair pathways, are also hypothesized to have a role in carcinogenesis.[78] The X-ray repair cross complementing one

(XRCC1) gene, especially the 194Trp allele, as well as a functional polymorphism of one of the *BER* genes, were related to thyroid cancer development and progression in Taiwan.[78] Moreover, gene polymorphism of the antioxidant enzyme GPX3 has also been analyzed; the C allele of rs8177412 was suggested to confer an increased risk for the development of differentiated thyroid cancer as compared to the T allele of rs8177412.[79]

6. Genetic predisposition to AITD

6.1. Susceptible loci in the human leukocyte antigen region

Genetic predisposition to AITD has been linked to a susceptible gene locus in the human leukocyte antigen (HLA) region and in non-HLA regions, including the cytotoxic T lymphocyte-associated molecule-4 (*CTLA-4)*, *CD40*, protein tyrosine phosphatase-22 (*PTPN22*), *TG*, and thyroid stimulating hormone receptor (*TSHR*) genes in Caucasians.[80] Linkage analysis revealed a positive association between Graves' disease (GD) and the HLA region in a Taiwanese population.[81] HLA-DR2 and DR9 were the first two associated alleles found.[82] HLA-B*4601, the predominant allele across Asian countries, was also reported to be a susceptible allele for both GD and Hashimoto thyroiditis (HT) in Taiwan.[83,84] Other dominant genes in different populations included HLA-A*0207 in adult GD patients and HLA-DRB1*0901 in children with GD.[83,85] More comprehensive HLA genotyping performed recently in Taiwan identified susceptible alleles with independent effects, including HLA-B*4601, HLA-DPB1*0501, DRB1*1502, and 1602.[84] Specifically, whereas DRB1*1202 and DQB1*0302 conferred a protective effect for GD, a high frequency of HLA-DRB1*0301 was detected in Caucasians with GD, and HLA-DPB1*0501 was shown to be the major allele associated with GD in Taiwan.[84]

6.2. CTLA-4 gene in GD

CTLA-4 functions as a T cell regulator, inhibiting T cell activation possibly through interfering with the costimulatory effect of CD28.[86] Meta-analysis of *CTLA-4* gene polymorphisms has showed a positive association of two SNPs, +49G>A and +6230G>A (CT60), with AITD.[86] In a Taiwanese population, the GG genotype and G allele of SNP CT60 was associated with susceptibility to GD in adults and children.[87,88] Besides the association with GD, the GG genotype and G allele of SNP +49G>A was also related to relapse of Graves' hyperthyroidism after withdrawal of antithyroid drugs.[89,90] Similar to the antigen presentation and T cell activation function of major histocompatibility complex (MHC), MHC class I-chain related gene A (*MICA*) functions as a stress-induced antigen and ligand for natural killer (NK) cell activation.[91] Microsatellite repeat polymorphism within the transmembrane region of *MICA* varied between individuals.[91] In children with GD, Lo et al[91] found that allele A5 was associated with higher risk for the development of GD.

6.3. Cytokine gene polymorphism and AITD

Association-related studies of cytokine gene polymorphism in AITD are discrepant.[92–100] In Taiwan, a positive association with GD was found in the allelic variants of the interleukin-1-β (*IL-1-β*) gene promoter (G-511C) and the tumor necrosis factor-α (*TNF-α*) gene promoter (G-238A and G-308A), but not the IL-1 receptor antagonist (*IL-1RA*) gene, the *IL-8* 3′ untranslated region (3′-UTR), or the *IL-4*, *IL-6*, and *IL-10* gene promoters.[93–96] In addition, a meta-analysis of eight studies by Lee et al[97] reported no association between *IL-4* gene SNPs and GD. However, a TA haplotype of the *IL-4* gene conferred risk for GD, suggesting that the haplotype-based method may be more powerful in studying gene–disease associations than individual SNPs.[97]

6.4. Other studies on HT

The number of genetic studies on HT was much lower than that of GD in Taiwan. In addition to the aforementioned positive association of HLA-B*4601 with HT, the C/G genotype and allele C of SNP rs187238 in the *IL-18* gene conferred an increased risk in children.[98] Analysis of four common polymorphisms of the vitamin-D receptor (*VDR*) gene, including FokI (rs10735810), TaqI (rs731236), BsmI (rs1544410), and ApaI (rs7975232), suggested that the homozygous C/C *VDR-FokI* gene polymorphism in exon 2 conferred a higher risk of developing HT in Chinese patients.[99] However, a recent meta-analysis suggested that the BsmI and TaqI polymorphisms were more strongly associated with AITD, whereas ApaI or FokI polymorphisms were not.[100]

In conclusion, the mutation spectrum of the Han-Chinese population observed in monogenic thyroid disorders was different from that of Caucasians. The single most common mutation in the *TPO* gene, c.2268insT, was observed in more than half of the cases of congenital hypothyroidism with *TPO* gene mutation in Taiwan. In the *PDS* gene, IVS7-2A>G (c.919-2A>G) accounted for > 80% of the mutations in the Han-Chinese population. All of the six mutations identified in the *TG* gene were novel, suggesting that there may be a distinct mutation spectrum in the Taiwanese population. By contrast, the epidemiology of different somatic mutations in differentiated thyroid cancer in Taiwan was similar to that of Western countries, with most of the mutations involving the MAPK pathway in a prevalent sequence of *BRAF* occurring more often than *RAS*, *RET*/PTC, and *TRK* mutations. In sporadic and MEN-associated MTC in Taiwan, two common mutation sites in the *RET* proto-oncogene at codons 634 and 918 were observed, which is consistent with results from other populations.

In genetic studies of AITDs, different dominant HLA alleles in Asians or Caucasians with GD were disclosed; HLA-DPB1*0501 was found in high frequency in Asian countries but not in the West. In addition, two SNPs of the *CTLA-4* gene, +49G>A and +6230G>A (CT60), were positively associated with GD in Taiwan. Analysis of cytokine genes in Taiwan as well as in other populations has been conflicting.

Thus, local genetic studies of thyroid disorders in Taiwan suggest that specific genetic patterns exist in some diseases in Han-Chinese whereas others were similar to the results observed worldwide.

References

1. Tsai WY, Lee JS, Chao MC, Chen LY, Lin SJ, Wu KH, et al. Prevalence of permanent primary congenital hypothyroidism in Taiwan. *J Formos Med Assoc* 1995;**94**:271−3.
2. Chen CY, Lee KT, Lee CT, Lai WT, Huang YB. Epidemiology and clinical characteristics of congenital hypothyroidism in an Asian population: a nationwide population-based study. *J Epidemiol* 2013;**23**:85−94.
3. Delange F. Neonatal screening for congenital hypothyroidism in Europe. Report of the Newborn Committee of the European Thyroid Association. *Acta Endocrinol Suppl (Copenh)* 1979;**223**:3−29.
4. Fisher DA, Dussault JH, Foley Jr TP, Klein AH, LaFranchi S, Larsen PR, et al. Screening for congenital hypothyroidism: results of screening one million North American infants. *J Pediatr* 1979;**94**:700−5.
5. Kopp P. Perspective: genetic defects in the etiology of congenital hypothyroidism. *Endocrinology* 2002;**143**:2019−24.
6. Park SM, Chatterjee VK. Genetics of congenital hypothyroidism. *J Med Genet* 2005;**42**:379−89.
7. Niu DM, Hwang B, Chu YK, Liao CJ, Wang PL, Lin CY. High prevalence of a novel mutation (2268 insT) of the thyroid peroxidase gene in Taiwanese patients with total iodide organification defect, and evidence for a founder effect. *J Clin Endocrinol Metab* 2002;**87**:4208−12.
8. Wu JY, Shu SG, Yang CF, Lee CC, Tsai FJ. Mutation analysis of thyroid peroxidase gene in Chinese patients with total iodide organification defect: identification of five novel mutations. *J Endocrinol* 2002;**172**:627−35.
9. Niu DM, Hsu JH, Chong KW, Huang CH, Lu YH, Kao CH, et al. Six new mutations of the thyroglobulin gene discovered in Taiwanese children presenting with thyroid dyshormonogenesis. *J Clin Endocrinol Metab* 2009;**94**:5045−52.
10. Dossena S, Nofziger C, Tamma G, Bernardinelli E, Vanoni S, Nowak C, et al. Molecular and functional characterization of human pendrin and its allelic variants. *Cell Physiol Biochem* 2011;**28**:451−66.
11. Persani L, Calebiro D, Cordella D, Weber G, Gelmini G, Libri D, et al. Genetics and phenomics of hypothyroidism due to TSH resistance. *Mol Cell Endocrinol* 2010;**322**:72−82.
12. Schoenmakers N, Moran C, Peeters RP, Visser T, Gurnell M, Chatterjee K. Resistance to thyroid hormone mediated by defective thyroid hormone receptor alpha. *Biochim Biophys Acta* 2013;**1830**:4004−8.
13. Mamanasiri S, Yesil S, Dumitrescu AM, Liao XH, Demir T, Weiss RE, et al. Mosaicism of a thyroid hormone receptor-beta gene mutation in resistance to thyroid hormone. *J Clin Endocrinol Metab* 2006;**91**:3471−7.
14. Endo Y, Onogi S, Umeki K, Yamamoto I, Kotani T, Ohtaki S, et al. Regional localization of the gene for thyroid peroxidase to human chromosome 2p25 and mouse chromosome 12C. *Genomics* 1995;**25**:760−1.
15. Kimura S, Kotani T, McBride OW, Umeki K, Hirai K, Nakayama T, et al. Human thyroid peroxidase: complete cDNA and protein sequence, chromosome mapping, and identification of two alternately spliced mRNAs. *Proc Natl Acad Sci USA* 1987;**84**:5555−9.
16. Niu DM, Lin CY, Hwang B, Jap TS, Liao CJ, Wu JY. Contribution of genetic factors to neonatal transient hypothyroidism. *Arch Dis Child Fetal Neonatal Ed* 2005;**90**:F69−72.
17. Coyle B, Coffey R, Armour JA, Gausden E, Hochberg Z, Grossman A, et al. Pendred syndrome (goitre and sensorineural hearing loss) maps to chromosome 7 in the region containing the nonsyndromic deafness gene DFNB4. *Nat Genet* 1996;**12**:421−3.
18. Sheffield VC, Kraiem Z, Beck JC, Nishimura D, Stone EM, Salameh M, et al. Pendred syndrome maps to chromosome 7q21-34 and is caused by an intrinsic defect in thyroid iodine organification. *Nat Genet* 1996;**12**:424−6.
19. Bizhanova A, Kopp P. Genetics and phenomics of Pendred syndrome. *Mol Cell Endocrinol* 2010;**322**:83−90.
20. Huang CJ, Lei TH, Chang WL, Tu TY, Shiao AS, Chiu CY, et al. A novel mutation in the *SLC26A4* gene in a Chinese family with Pendred syndrome. *Int J Pediatr Otorhinolaryngol* 2013;**77**:1495−9.
21. Pourova R, Janousek P, Jurovcik M, Dvorakova M, Malikova M, Raskova D, et al. Spectrum and frequency of SLC26A4 mutations among Czech patients with early hearing loss with and without Enlarged Vestibular Aqueduct (EVA). *Ann Hum Genet* 2010;**74**:299−307.
22. Yang JJ, Tsai CC, Hsu HM, Shiao JY, Su CC, Li SY. Hearing loss associated with enlarged vestibular aqueduct and Mondini dysplasia is caused by splice-site mutation in the *PDS* gene. *Hear Res* 2005;**199**:22−30.
23. Lai CC, Chiu CY, Shiao AS, Tso YC, Wu YC, Tu TY, et al. Analysis of the *SLC26A4* gene in patients with Pendred syndrome in Taiwan. *Metabolism* 2007;**56**:1279−84.
24. Mendive FM, Rivolta CM, Moya CM, Vassart G, Targovnik HM. Genomic organization of the human thyroglobulin gene: the complete intron-exon structure. *Eur J Endocrinol* 2001;**145**:485−96.
25. Rivolta CM, Targovnik HM. Molecular advances in thyroglobulin disorders. *Clin Chim Acta* 2006;**374**:8−24.
26. Kopp P. The TSH receptor and its role in thyroid disease. *Cell Mol Life Sci* 2001;**58**:1301−22.
27. Narumi S, Muroya K, Abe Y, Yasui M, Asakura Y, Adachi M, et al. TSHR mutations as a cause of congenital hypothyroidism in Japan: a population-based genetic epidemiology study. *J Clin Endocrinol Metab* 2009;**94**:1317−23.
28. Chang WC, Liao CY, Chen WC, Fan YC, Chiu SJ, Kuo HC, et al. R450H TSH receptor mutation in congenital hypothyroidism in Taiwanese children. *Clin Chim Acta* 2012;**413**:1004−7.
29. Tang KT, Yang HJ, Choo KB, Lin HD, Fang SL, Braverman LE. A point mutation in the albumin gene in a Chinese patient with familial dysalbuminemic hyperthyroxinemia. *Eur J Endocrinol* 1999;**141**:374−8.
30. Su CC, Wu YC, Chiu CY, Won JG, Jap TS. Two novel mutations in the gene encoding thyroxine-binding globulin (TBG) as a cause of complete TBG deficiency in Taiwan. *Clin Endocrinol* 2003;**58**:409−14.
31. Rajatanavin R, Fournier L, DeCosimo D, Abreau C, Braverman LE. Elevated serum free thyroxine by thyroxine analog radioimmunoassays in euthyroid patients with familial dysalbuminemic hyperthyroxinemia. *Ann Intern Med* 1982;**97**:865−6.
32. Stockigt JR, Topliss DJ, Barlow JW, White EL, Hurley DM, Taft P. Familial euthyroid thyroxine excess: an appropriate response to abnormal thyroxine binding associated with albumin. *J Clin Endocrinol Metab* 1981;**53**:353−9.
33. Minghetti PP, Ruffner DE, Kuang WJ, Dennison OE, Hawkins JW, Beattie WG, et al. Molecular structure of the human albumin gene is revealed by nucleotide sequence within q11-22 of chromosome 4. *J Biol Chem* 1986;**261**:6747−57.
34. Petersen CE, Scottolini AG, Cody LR, Mandel M, Reimer N, Bhagavan NV. A point mutation in the human serum albumin gene results in familial dysalbuminaemic hyperthyroxinaemia. *J Med Genet* 1994;**31**:355−9.
35. Wada N, Chiba H, Shimizu C, Kijima H, Kubo M, Koike T. A novel missense mutation in codon 218 of the albumin gene in a distinct phenotype of familial dysalbuminemic hyperthyroxinemia in a Japanese kindred. *J Clin Endocrinol Metab* 1997;**82**:3246−50.
36. Mori Y, Miura Y, Oiso Y, Hisao S, Takazumi K. Precise localization of the human thyroxine-binding globulin gene to chromosome Xq22.2 by fluorescence *in situ* hybridization. *Hum Genet* 1995;**96**:481−2.
37. Flink IL, Bailey TJ, Gustafson TA, Markham BE, Morkin E. Complete amino acid sequence of human thyroxine-binding globulin deduced from cloned DNA: close homology to the serine antiproteases. *Proc Natl Acad Sci USA* 1986;**83**:7708−12.
38. Takamatsu J, Refetoff S, Charbonneau M, Dussault JH. Two new inherited defects of the thyroxine-binding globulin (TBG) molecule presenting as partial TBG deficiency. *J Clin Invest* 1987;**79**:833−40.

39. Mori Y, Jing P, Kayama M, Fujieda K, Hasegawa T, Nogimori T, et al. Gene amplification as a common cause of inherited thyroxine-binding globulin excess: analysis of one familial and two sporadic cases. *Endocr J* 1999;**46**:613–9.
40. Miura Y, Kambe F, Yamamori I, Mori Y, Tani Y, Murata Y, et al. A truncated thyroxine-binding globulin due to a frameshift mutation is retained within the rough endoplasmic reticulum: a possible mechanism of complete thyroxine-binding globulin deficiency in Japanese. *J Clin Endocrinol Metab* 1994;**78**:283–7.
41. Mori Y, Takeda K, Charbonneau M, Refetoff S. Replacement of Leu227 by Pro in thyroxine-binding globulin (TBG) is associated with complete TBG deficiency in three of eight families with this inherited defect. *J Clin Endocrinol Metab* 1990;**70**:804–9.
42. Hundahl SA, Fleming ID, Fremgen AM, Menck HR. A National Cancer Data Base report on 53,856 cases of thyroid carcinoma treated in the U.S., 1985–1995. *Cancer* 1998;**83**:2638–48.
43. Chiacchio S, Lorenzoni A, Boni G, Rubello D, Elisei R, Mariani G. Anaplastic thyroid cancer: prevalence, diagnosis and treatment. *Minerva Endocrinol* 2008;**33**:341–57.
44. Xing M. Molecular pathogenesis and mechanisms of thyroid cancer. *Nat Rev Cancer* 2013;**13**:184–99.
45. Haraldsdottir S, Shah MH. An update on clinical trials of targeted therapies in thyroid cancer. *Curr Opin Oncol* 2014;**26**:36–44.
46. MacCorkle RA, Tan TH. Mitogen-activated protein kinases in cell-cycle control. *Cell Biochem Biophys* 2005;**43**:451–61.
47. Tang KT, Lee CH. BRAF mutation in papillary thyroid carcinoma: pathogenic role and clinical implications. *J Chin Med Assoc* 2010;**73**:113–28.
48. Santoro M, Carlomagno F, Hay ID, Herrmann MA, Grieco M, Melillo R, et al. Ret oncogene activation in human thyroid neoplasms is restricted to the papillary cancer subtype. *J Clin Invest* 1992;**89**:1517–22.
49. Greco A, Pierotti MA, Bongarzone I, Pagliardini S, Lanzi C, Della Porta G. TRK-T1 is a novel oncogene formed by the fusion of TPR and TRK genes in human papillary thyroid carcinomas. *Oncogene* 1992;**7**:237–42.
50. Lee CH, Hsu LS, Chi CW, Chen GD, Yang AH, Chen JY. High frequency of rearrangement of the RET protooncogene (RET/PTC) in Chinese papillary thyroid carcinomas. *J Clin Endocrinol Metab* 1998;**83**:1629–32.
51. Liu RT, Chou FF, Wang CH, Lin CL, Chao FP, Chung JC, et al. Low prevalence of RET rearrangements (RET/PTC1, RET/PTC2, RET/PTC3, and ELKS-RET) in sporadic papillary thyroid carcinomas in Taiwan Chinese. *Thyroid* 2005;**15**:326–35.
52. Learoyd DL, Messina M, Zedenius J, Guinea AI, Delbridge LW, Robinson BG. RET/PTC and RET tyrosine kinase expression in adult papillary thyroid carcinomas. *J Clin Endocrinol Metab* 1998;**83**:3631–5.
53. Chung JH, Hahm JR, Min YK, Lee MS, Lee MK, Kim KW, et al. Detection of RET/PTC oncogene rearrangements in Korean papillary thyroid carcinomas. *Thyroid* 1999;**9**:1237–43.
54. Jhiang SM, Caruso DR, Gilmore E, Ishizaka Y, Tahira T, Nagao M, et al. Detection of the PTC/retTPC oncogene in human thyroid cancers. *Oncogene* 1992;**7**:1331–7.
55. Kuo CS, Lin CY, Hsu CW, Lee CH, Lin HD. Low frequency of rearrangement of TRK protooncogene in Chinese thyroid tumors. *Endocrine* 2000;**13**:341–4.
56. Liu RT, Hou CY, You HL, Huang CC, Hock L, Chou FF, et al. Selective occurrence of ras mutations in benign and malignant thyroid follicular neoplasms in Taiwan. *Thyroid* 2004;**14**:616–21.
57. Liu RT, Chen YJ, Chou FF, Li CL, Wu WL, Tsai PC, et al. No correlation between BRAFV600E mutation and clinicopathological features of papillary thyroid carcinomas in Taiwan. *Clin Endocrinol* 2005;**63**:461–6.
58. Nikiforova MN, Kimura ET, Gandhi M, Biddinger PW, Knauf JA, Basolo F, et al. BRAF mutations in thyroid tumors are restricted to papillary carcinomas and anaplastic or poorly differentiated carcinomas arising from papillary carcinomas. *J Clin Endocrinol Metab* 2003;**88**:5399–404.
59. Carta C, Moretti S, Passeri L, Barbi F, Avenia N, Cavaliere A, et al. Genotyping of an Italian papillary thyroid carcinoma cohort revealed high prevalence of BRAF mutations, absence of RAS mutations and allowed the detection of a new mutation of BRAF oncoprotein (BRAF(V599lns)). *Clin Endocrinol* 2006;**64**:105–9.
60. Kimura ET, Nikiforova MN, Zhu Z, Knauf JA, Nikiforov YE, Fagin JA. High prevalence of BRAF mutations in thyroid cancer: genetic evidence for constitutive activation of the RET/PTC-RAS-BRAF signaling pathway in papillary thyroid carcinoma. *Cancer Res* 2003;**63**:1454–7.
61. Soares P, Trovisco V, Rocha AS, Lima J, Castro P, Preto A, et al. BRAF mutations and RET/PTC rearrangements are alternative events in the etiopathogenesis of PTC. *Oncogene* 2003;**22**:4578–80.
62. Kim TH, Park YJ, Lim JA, Ahn HY, Lee EK, Lee YJ, et al. The association of the BRAF(V600E) mutation with prognostic factors and poor clinical outcome in papillary thyroid cancer: a meta-analysis. *Cancer* 2012;**118**:1764–73.
63. Chou CK, Chen RF, Chou FF, Chang HW, Chen YJ, Lee YF, et al. miR-146b is highly expressed in adult papillary thyroid carcinomas with high risk features including extrathyroidal invasion and the BRAF(V600E) mutation. *Thyroid* 2010;**20**:489–94.
64. Chou CK, Yang KD, Chou FF, Huang CC, Lan YW, Lee YF, et al. Prognostic implications of miR-146b expression and its functional role in papillary thyroid carcinoma. *J Clin Endocrinol Metab* 2013;**98**:E196–205.
65. Ho YS, Tseng SC, Chin TY, Hsieh LL, Lin JD. *p53* gene mutation in thyroid carcinoma. *Cancer Lett* 1996;**103**:57–63.
66. Hsieh MC, Lin SF, Shin SJ, Liu TC, Chang JG, Lee JP. Mutation analysis of PTEN/MMAC 1 in sporadic thyroid tumors. *Kaohsiung J Med Sci* 2000;**16**:9–12.
67. Chang TJ, Tsai TC, Wu YL, Yang HM, Chi CW, Yang AH, et al. Abnormal transcripts of *FHIT* gene in thyroid cancer. *Oncol Rep* 1998;**5**:245–7.
68. Nag S, Qin J, Srivenugopal KS, Wang M, Zhang R. The MDM2-p53 pathway revisited. *J Biomed Res* 2013;**27**:254–71.
69. Ngeow J, Mester J, Rybicki LA, Ni Y, Milas M, Eng C. Incidence and clinical characteristics of thyroid cancer in prospective series of individuals with Cowden and Cowden-like syndrome characterized by germline PTEN, SDH, or KLLN alterations. *J Clin Endocrinol Metab* 2011;**96**:E2063–71.
70. Wang HM, Huang YW, Huang JS, Wang CH, Kok VC, Hung CM, et al. Anaplastic carcinoma of the thyroid arising more often from follicular carcinoma than papillary carcinoma. *Ann Surg Oncol* 2007;**14**:3011–8.
71. Ganeshan D, Paulson E, Duran C, Cabanillas ME, Busaidy NL, Charnsangavej C. Current update on medullary thyroid carcinoma. *AJR Am J Roentgenol* 2013;**201**:867–76.
72. Ceolin L, Siqueira DR, Romitti M, Ferreira CV, Maia AL. Molecular basis of medullary thyroid carcinoma: the role of RET polymorphisms. *Int J Mol Sci* 2012;**13**:221–39.
73. Huang CN, Wu SL, Chang TC, Huang SH, Chang TJ. RET proto-oncogene mutations in patients with apparently sporadic medullary thyroid carcinoma. *J Formos Med Assoc* 1998;**97**:541–6.
74. Chang CF, Yang WS, Su YN, Wu IL, Chang TC. Mutational spectrum of multiple endocrine neoplasia type 2 and sporadic medullary thyroid carcinoma in Taiwan. *J Formos Med Assoc* 2009;**108**:402–8.
75. Chang TC, Wu SL, Hsiao YL. Medullary thyroid carcinoma: pitfalls in diagnosis by fine needle aspiration cytology and relationship of cytomorphology to RET proto-oncogene mutations. *Acta Cytol* 2005;**49**:477–82.
76. Lin JD, Chao TC. Vascular endothelial growth factor in thyroid cancers. *Cancer Biother Radiopharm* 2005;**20**:648–61.
77. Hsiao PJ, Lu MY, Chiang FY, Shin SJ, Tai YD, Juo SH. Vascular endothelial growth factor gene polymorphisms in thyroid cancer. *J Endocrinol* 2007;**195**:265–70.
78. Chiang FY, Wu CW, Hsiao PJ, Kuo WR, Lee KW, Lin JC, et al. Association between polymorphisms in DNA base excision repair genes XRCC1, APE1, and ADPRT and differentiated thyroid carcinoma. *Clin Cancer Res* 2008;**14**:5919–24.

79. Lin JC, Kuo WR, Chiang FY, Hsiao PJ, Lee KW, Wu CW, et al. Glutathione peroxidase 3 gene polymorphisms and risk of differentiated thyroid cancer. *Surgery* 2009;**145**:508–13.
80. Jacobson EM, Tomer Y. The genetic basis of thyroid autoimmunity. *Thyroid* 2007;**17**:949–61.
81. Chen PL, Fann CS, Chang CC, Wu IL, Chiu WY, Lin CY, et al. Linkage of Graves' disease to the human leucocyte antigen region in the Chinese-Han population in Taiwan. *Clin Endocrinol* 2007;**66**:646–51.
82. Tsai KS, Hsieh RP, Chang CC, Chen FW, Lee SC. Association of HLA-DR tissue types with Graves' disease in Taiwan. *Taiwan Yi Xue Hui Za Zhi* 1989;**88**:336–41.
83. Huang SM, Wu TJ, Lee TD, Yang EK, Shaw CK, Yeh CC. The association of HLA -A, -B, and -DRB1 genotypes with Graves' disease in Taiwanese people. *Tissue Antigens* 2003;**61**:154–8.
84. Chen PL, Fann CS, Chu CC, Chang CC, Chang SW, Hsieh HY, et al. Comprehensive genotyping in two homogeneous Graves' disease samples reveals major and novel HLA association alleles. *PLoS One* 2011;**6**:e16635.
85. Wu YL, Chang TY, Chu CC, Huang CY, Lo FS, Ting WH, et al. The *HLA-DRB1* gene and Graves disease in Taiwanese children: a case-control and family-based study. *Tissue Antigens* 2012;**80**:224–30.
86. Kavvoura FK, Akamizu T, Awata T, Ban Y, Chistiakov DA, Frydecka I, et al. Cytotoxic T-lymphocyte associated antigen 4 gene polymorphisms and autoimmune thyroid disease: a meta-analysis. *J Clin Endocrinol Metab* 2007;**92**:3162–70.
87. Weng YC, Wu MJ, Lin WS. CT60 single nucleotide polymorphism of the CTLA-4 gene is associated with susceptibility to Graves' disease in the Taiwanese population. *Ann Clin Lab Sci* 2005;**35**:259–64.
88. Tsai ST, Huang CY, Lo FS, Chang YT, Tanizawa T, Chen CK, et al. Association of CT60 polymorphism of the *CTLA4* gene with Graves' disease in Taiwanese children. *J Pediatr Endocrinol Metab* 2008;**21**:665–72.
89. Wang PW, Liu RT, Juo SH, Wang ST, Hu YH, Hsieh CJ, et al. Cytotoxic T lymphocyte-associated molecule-4 polymorphism and relapse of Graves' hyperthyroidism after antithyroid withdrawal. *J Clin Endocrinol Metab* 2004;**89**:169–73.
90. Wang PW, Chen IY, Liu RT, Hsieh CJ, Hsi E, Juo SH. Cytotoxic T lymphocyte-associated molecule-4 gene polymorphism and hyperthyroid Graves' disease relapse after antithyroid drug withdrawal: a follow-up study. *J Clin Endocrinol Metab* 2007;**92**:2513–8.
91. Lo FS, Lee YJ, Huang CY, Lin CH, Chang SC, Dang CW, et al. Polymorphism in the transmembrane region of the major histocompatibility complex class I chain-related gene A: association of five GCT repetitions with Graves' disease in children. *Thyroid* 2003;**13**:839–43.
92. Hunt PJ, Marshall SE, Weetman AP, Bell JI, Wass JA, Welsh KI. Cytokine gene polymorphisms in autoimmune thyroid disease. *J Clin Endocrinol Metab* 2000;**85**:1984–8.
93. Chen RH, Chen WC, Chang CT, Tsai CH, Tsai FJ. Interleukin-1-beta gene, but not the interleukin-1 receptor antagonist gene, is associated with Graves' disease. *J Clin Lab Anal* 2005;**19**:133–8.
94. Chen RH, Chen WC, Wang TY, Tsai CH, Tsai FJ. Lack of association between pro-inflammatory cytokine (IL-6, IL-8 and TNF-alpha) gene polymorphisms and Graves' disease. *Int J Immunogenet* 2005;**32**:343–7.
95. Chen RH, Chang CT, Wang TY, Chen CC, Tsai CH, Tsai FJ. Lack of association between interleukin-4 gene polymorphisms and autoimmune thyroid diseases amongst Taiwanese Chinese. *Endocrine* 2007;**32**:170–4.
96. Shiau MY, Huang CN, Yang TP, Hwang YC, Tsai KJ, Chi CJ, et al. Cytokine promoter polymorphisms in Taiwanese patients with Graves' disease. *Clin Biochem* 2007;**40**:213–7.
97. Lee YJ, Huang CY, Ting WH, Lee HC, Guo WL, Chen WF, et al. Association of an IL-4 gene haplotype with Graves disease in children: experimental study and meta-analysis. *Hum Immunol* 2011;**72**:256–61.
98. Huang CY, Ting WH, Lo FS, Wu YL, Chang TY, Chan HW, et al. The IL18 gene and Hashimoto thyroiditis in children. *Hum Immunol* 2013;**74**:120–4.
99. Lin WY, Wan L, Tsai CH, Chen RH, Lee CC, Tsai FJ. Vitamin D receptor gene polymorphisms are associated with risk of Hashimoto's thyroiditis in Chinese patients in Taiwan. *J Clin Lab Anal* 2006;**20**:109–12.
100. Feng M, Li H, Chen SF, Li WF, Zhang FB. Polymorphisms in the vitamin D receptor gene and risk of autoimmune thyroid diseases: a meta-analysis. *Endocrine* 2013;**43**:318–26.