Thyroid disorders include autoimmune thyroid diseases (AITD), thyroid goiter, nodule and cancer. AITD mainly consist of autoimmune thyroiditis and Graves disease. The common characteristic of thyroid disorders is female preponderance in their prevalence. The female-to-male rate ratio is reported at 4~6:1 for AITD and about 3~4:1 for thyroid nodule. For PTC, it is greatest during reproductive age and drops from five and more in patients aged 20-24, to 3.4 in patients aged 35-44 to one in patients over 80. The effects of female gonadal hormones and X chromosome inactivation on thyroid gland and immune system greatly contribute to the female predilection of AITD. The former mainly include prolactin and estrogen. The direct actions of estrogen on the thyroid tissue contribute to the development of thyroid goiter, nodule and cancer in women.

**Key words:** Thyroid - Autoimmunity - Neoplasms - Estrogens.

Thyroid disorders have become more and more common in the general population, including mainly autoimmune thyroid diseases (AITD), thyroid nodule and cancer. AITD consist of autoimmune thyroiditis (AIT) and Graves disease (GD). Their prevalence is approximately 5% and 1% in adults, respectively. The most common type of thyroid cancer is papillary thyroid cancer (PTC). According to the epidemiological studies, both AITD and PTC are more prevalent in women than in men. The female-to-male rate ratio of AITD is reported at 4~6:1 in the general population. For PTC, it is greatest during reproductive age and drops from five and more in patients aged 20-24, to 3.4 in patients aged 35-44 to one in patients over 80. Thyroid nodule is the most frequent endocrine neoplasm. Its female-to-male rate ratio is about 3~4:1. Goiter is also more common in women than men even in iodine-sufficient areas. In addition, the incidence of AIT is about 4 times higher in women than men. Polycystic ovary syndrome (PCOS) is also a very common endocrine disease among women at reproductive age. Its incidence rate is about 6-10%. PCOS has been found not only to be associated with higher prevalence of serum positive thyroid autoantibodies in general population, but also increase the risk for thyroid disorders. It has been reported that high estrogen-to-progesterone ratio owing
to anovulatory cycles in the patients with PCOS may modulate immune response and contribute to the development of AITD.\textsuperscript{9} AITD also spontaneously remiss during pregnancy and aggravate after delivery.\textsuperscript{3, 10} These findings have suggested the potential effects of female gonadal hormones and X chromosome inactivation on thyroid gland and immune system.\textsuperscript{3, 10}

This paper is a brief review on the reported influences of prolactin (PRL), estrogen and X chromosome inactivation on thyroid disorders for better understanding their sex dichotomy.

**The effects of prolactin on thyroid disorders**

PRL is one of the most prominent immunostimulatory hormones in the immuno-neuro-endocrine network since 1980, which has been known to interfere with the induction of B cell tolerance, enhance the proliferation of lymphocytes, and promote the production of cytokines and autoantibodies.\textsuperscript{11, 12} It is a polypeptide hormone that produced by the pituitary gland and other cells, particularly lymphocytes. Hyperprolactinemia has been found not only in patients with systemic lupus erythematosus (SLE),\textsuperscript{13} but also in those with AIT.\textsuperscript{14} The impact of PRL on the immune system has exhibited a biphasic pattern. At physiological level, PRL is trophic for the lymphocytes, while its insufficiency or hypersecretion has been found to be involved in the development of autoimmune diseases.\textsuperscript{15} Several studies have focused on the relationship between the level of PRL and thyroid disorders. The prevalence of AITD, thyroid nodule and goiter has all been reported to be significantly increased in patients with hyperprolactinaemia.\textsuperscript{16} Ferrari et al. observed that the prevalence of thyroid autoantibody positivity was significantly higher in patients with prolactinoma than the control groups.\textsuperscript{17} Poyraz et al. investigated the occurrence of AIT in patients with antipsychotic-induced hyperprolactinemia who had no history of obvious thyroid disease.\textsuperscript{18} They found that the prevalence of hyperprolactinemia was significantly higher in patients with positive thyroid autoantibodies than in those with negative values.\textsuperscript{18} In the subgroup analysis for gender, the production of thyroid autoantibodies in women seemed to be related to increased levels of PRL.\textsuperscript{18} They found that long-term antipsychotic treatment can induce thyroid autoimmunity due to enhanced PRL secretion.\textsuperscript{18}

Graves’ disease (GD) is another type of AITD. It usually aggravates or relapses in the postpartum period. We have found that PRL could affect the \textit{in-vitro} interactions between peripheral blood mononuclear cells and autologous thyrocytes prepared from GD patients, and may eventually influence the shift of helper T (Th) cells.\textsuperscript{19} In addition, PRL can exert indirect effects on CD40 expressions on the surfaces of thyrocytes by antagonizing the modulatory actions of IFN-\gamma and IL-4 with dose-related effects.\textsuperscript{20} PRL-like substance was detectable in GD thyroid tissue and mainly distributed in infiltrating mononuclear cells and vascular endothelial cells adjacent to the infiltrates.\textsuperscript{21} It was also present in both thyroid follicular cells nearby the infiltrates and the local connective tissue.\textsuperscript{21} The percentages of B cells, the detection rates and titers of thyroid autoantibodies (TMAb or TPOAb and TgAb) were significantly increased in peripheral blood of hyperprolactinemic patients as compared with the general population.\textsuperscript{22} Very small amounts of specific PRL binding to thyroid membranes have been detected.\textsuperscript{18} These indicate that PRL could affect the development of GD and a high PRL level may lead to the aggravation or occurrence of AITD.

**The effects of estrogen on thyroid disorders**

Estrogen is not only the predominant sex hormone in females, but it also stimulates the secretion of PRL from the pituitary gland. Estrogen can exert multiple regulation of the immune response.\textsuperscript{23} The growth, differentiation and proliferation of
lymphocytes, secretion of cytokines and production of antibodies are typically influenced by estrogens.\textsuperscript{23-25} Estrogen receptors (ERs) mainly include two subtypes - ER-\(\alpha\) and ER-\(\beta\).\textsuperscript{26} Both subtypes are widely distributed in the immune system.\textsuperscript{23} It has been well known that ER-\(\alpha\) activation mediates the stimulatory effects of estrogen on the development of lupus erythematusus, a systemic autoimmune disease,\textsuperscript{27} while ER-\(\beta\) plays a key role in the inhibitory actions of estrogen on experimental autoimmune encephalomyelitis, an organ-specific autoimmune disease.\textsuperscript{28} In an earlier study, estradiol treatment significantly increased the production of Tg autoantibody (TgAb) in sham-operated and castrated male mice when experimental autoimmune thyroiditis (EAT) was induced.\textsuperscript{29} Guo et al. have reported that the development of EAT were severely enhanced in female SD rats after ovariectomy (OVX).\textsuperscript{30} The findings suggest that female sex steroid-estrogen may play a stimulatory role in the pathogenesis of AIT, although the individual roles of ER subtypes are still unknown. CBA/J mice are the commonly-used animals for establishment of EAT by immunization with Tg and Freund’s adjuvant, which also exhibits a female predilection of the disease. In our recent study, administration of ER-\(\beta\)-specific agonist-diarylpropionitrile (DPN) significantly decreased the percentage of Tregs (CD4\(^+\)CD25\(^++\)Foxp3\(^+\)) in CD4\(^+\)T cells, that of GITR-expressing cells in Tregs and the mean fluorescence intensity of GITR in the spleen of female CBA/J mice, which indicates the function of splenic Tregs can be suppressed due to ER\(\beta\) activation and autoimmune responses may be promoted.\textsuperscript{31} In addition, 2-methoxyestriadiol (2-ME) is one of the main endogenous estrogen metabolites present in the sera of women. The \textit{in-vitro} experiment has shown that 2-ME exposure disrupted the structural integrity of cultured thyroid follicles, caused cell apoptosis and resulted in a higher release of the autoantigen-thyroid peroxidase (TPO), which may contribute to a higher incidence ofAITD in females.\textsuperscript{32}

Since ER expression was found in the neoplastic and non-neoplastic thyroid glands about 30 years ago, thyroid has been considered as one of the non-classical target organs for the female sex steroid.\textsuperscript{33} ER-\(\alpha\) and ER-\(\beta\) expressions in thyroid tissue and cell lines have been investigated at both mRNA and protein levels. In addition, the splice variants of ER-\(\beta\) (ER-\(\beta\)\(_1\)-ER-\(\beta\)5) with truncations or insertions in the C-terminal ligand-binding domain have been reported in several tissues. The expressions of both the two ER subtypes have been found in benign and malignant thyroid tissues, although the findings are discordant.\textsuperscript{33, 34} Age, gender or tumor types used to exist as confounding factors in their statistical analyses.\textsuperscript{33, 34} We have recently explored the ER subtype expression in female patients with PTC and nodular thyroid goiter (NTG) stratified by age.\textsuperscript{34, 35} We found that ER-\(\alpha\) expression was significantly increased in female PTC patients of reproductive age (18-45 years old) as compared with that of age-matched NTG women, whereas ER-\(\beta\)1 was shown in an opposite pattern.\textsuperscript{34} No difference was found in ER-\(\alpha\) or ER\(\beta\)1 expression pattern between female PTC patients of reproductive age and those of advanced reproductive age (>45 years old).\textsuperscript{34} The above findings suggest that ER-\(\alpha\) may mediate the stimulatory effect of estrogen on the occurrence of PTC. We further compared the difference between expression pattern of ER-\(\beta\)1 (wild-type ER-\(\beta\)) and that of ER-\(\beta\)2 (ER-\(\beta\)cx, a c-terminal truncated splice variant) in PTC and NTG tissues. ER-\(\beta\)2 preferentially forms a heterodimer with ER-\(\alpha\) and inhibits DNA binding by ER-\(\alpha\). Our study showed that both nuclear and nucleocytoplasmic ER\(\beta\)1 expressions were significantly lower in PTC lesions than NTG tissue, while ER-\(\beta\)2 expression was significantly higher in the former than the latter. There was no significant difference in ER-\(\beta\)2 expression between reproductive-aged and advanced reproductive-aged female patients with PTC.\textsuperscript{35} Thus, the balance of ER-\(\alpha\)/ER-\(\beta\) expression seems to be involved in the development of PTC in female patients of reproductive age. The activation of ER by estrogen has been found to modulate the biological behaviors
of PTC, such as proliferation, growth and metastasis. An in-vitro study has found that estradiol treatment caused a significant increase in the relative thyroid weights of female rats. The activity of TPO was significantly enhanced in intact adult rats exposed to estradiol. Since serum TSH was not significantly altered, the above results indicate a direct effect of estradiol on the thyroid tissue. Using a rat two-stage thyroid tumorigenesis model, Son et al. has also found that estradiol exposure significantly increased thyroid weights and incidence of thyroid proliferative lesions in both female and male rats, and even induced a follicular carcinoma in a male rat. The in-vitro experiments have not only further demonstrated the above in-vivo findings, but also explored the related mechanisms for estrogen's actions. It has been found that estradiol enhanced the growth of ER-α-expressing FRTL-5 cells in either the absence or presence of TSH, and decreased the expression of the sodium/iodide symporter (NIS) induced by TSH, which may contribute to the female predilection of thyroid goiter. In addition, estradiol enhanced the proliferation of human WRO, FRO, and ARO thyroid carcinoma cells through up-regulation of c-fos, cyclin A, and D1 expressions, indicating the potential molecular mechanisms for estrogen to stimulate cancer progression. A recent study has found that thyroid stem and progenitor cells can express both ER subtypes with eight times higher levels of ER-α than that of the differentiated thyrocytes. Estradiol can stimulate the growth of thyroid stem/progenitor cells and suppress TSH-induced differentiation, which may lead to the formation of hypofunctioning or non-functioning thyroid nodules in females. Furthermore, the individual roles of ER subtypes in the pathogenesis of thyroid cancer have been investigated. Zeng et al. have reported that ER-α may mediate the stimulatory actions on the proliferation of thyroid cancer cells by estrogen through up-regulation of Bcl-2 and down-regulation of Bax expression level in an ERK1/2-related pathway. Our recent study has also shown that BCPAP cells (a typical PTC cell line) express both ER-α and ER-β. Estradiol and PPT (a potent ER-α-selective agonist) can promote while DPN (a potent ER-β-selective agonist) inhibit the migration and invasion of BCPAP cells in an in-vitro experimental model system that is modulated by E-cadherin, vimentin, and MMP-9, indicating that ER-α and ER-β play a differential role in the modulation of PTC metastasis. However, the relationship between female sex steroid hormones and thyroid cancer still await further investigation.

The effects of X chromosome mosaicism on thyroid disorders

In addition to the effects of sex hormones, genetic traits are associated with autoimmunity, including pregnancy related microchimerism and skewed X chromosome inactivation (XCI) in the females. Under normal conditions, one of two X chromosomes is randomly inactivated due to methylation in a female cell. However, disturbances in XCI mosaicism may be related to the development of autoimmune diseases. Autoimmunity is usually inhibited by central and peripheral immune tolerance established through exposure of self-antigens in the thymus and to the autoreactive T cells. XCI disturbances may result in the lack of exposure to self-antigens in the thymus, and eventually promote the occurrence of AITD. A recent meta-analysis has shown that the odds ratio of significantly skewed XCI was 2.54 (95% CI 1.58-4.10) for GD and 2.40 (95% CI 1.10-5.26) for AIT, which support its role in female preponderance in AITD. Skewed XCI has been considered as a epigenetic factor affecting the genetic predisposition of the females to autoimmunity. Some subsequent events, such as exposure to microbes, sex hormones, or environmental endocrine interrupters, may trigger a cascade response towards AITD.

Altogether, one common feature for most of the thyroid disorders is female preponderance in their prevalence. More and more studies have indicated the important roles of female gonadal hormones and X chro-
mosome inactivation in the pathogenesis of thyroid diseases. Further investigation of related mechanisms may help to develop new therapeutic and prophylactic strategies for the diseases.

References


