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# Are thyroid nodules associated with sex-related hormones? A cross-sectional SPECT-China study

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**Objective** Little is known about the association between thyroid nodules (TNs) and endogenous sex hormones. We aimed to investigate the relationship between TNs and sex-related hormones among men in China.

**Setting** The data were obtained from a cross-sectional study Survey on Prevalence in East China for Metabolic Diseases and Risk Factors (SPECT-China study, 2014–2015) based on the population.

**Participants** In total, 4024 men over 18 years of age who were not using hormone replacement therapy and who underwent complete assays of the serum total testosterone (T), oestradiol ($E_2$), follicle-stimulating hormone (FSH), luteinising hormone (LH) and sex hormone-binding globulin (SHBG) levels as well as thyroid ultrasonography (US) enrolled in this study.

**Results** Of the 4024 participants (54.15±13.08 years old), 1667 participants (41.4%) had TNs. Men with TN(s) (TN(+) group) had significantly lower levels of total T and SHBG and higher $E_2$/T levels compared with the men without TN(s) (TN(–) group) ($p<0.05$). The TN prevalence decreased with the quartiles of the SHBG level ($p<0.05$). Binary logistic analysis showed that lower quartiles of SHBG had a greater risk of TN(s) (all p for trend <0.05). This association persisted in the fully adjusted model (p for trend=0.017), in which, for the lowest compared with the highest quartile of SHBG, the OR of TN(s) was 1.42 (95% CI 1.07 to 1.89). No statistically significant association was found between sex-related hormones and US characteristics associated with malignancy (nodule >10 mm, microcalcification and a 'taller' than 'wider' shape).

**Conclusions** TNs are highly prevalent in men in China. A lower SHBG level was significantly associated with TN among men. The potential role of SHBG in the pathogenesis of the TN remains to be elucidated.

## Strengths and limitations of this study

- ► This is the first study to evaluate the relationship of sex hormones and thyroid nodules (TNs) in a relatively large sample of men in China.
- ► Anthropometric measurements and questionnaires were completed by the same trained research group, and all thyroid ultrasound examinations were performed by the same two doctors with strong quality control.
- ► Survey on Prevalence in East China for Metabolic Diseases and Risk Factors (SPECT China) study was performed in a general population instead of a clinic-based population, making the results potentially more generalisable.
- ► Because of the cross-sectional nature of the study, no causal inferences can be drawn.
- ► The gold standard to diagnose a TN is biopsy, and the use of thyroid ultrasonography has certain limitations.

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## INTRODUCTION

With changes in the medical practice, particularly the increased use of ultrasonography (US) and fine-needle aspiration biopsy, the incidence of a thyroid nodule (TN), even thyroid cancer, is increasing worldwide.[1] Currently, the estimated prevalence based on ultrasound ranges from 13% to 67% in the general population.[2] The prevalence of TNs is high even in healthy adults in China at approximately 30% to 50%.[3–5] Never in history has the issue of TNs aroused such great concern in the Chinese public as at present.

The American Association of Clinical Endocrinologists (AACE)/Associazione Medici Endocrinologi (AME)/European Thyroid Association (ETA) Thyroid Nodule Guidelines[6] declare that TNs are more common in elderly persons, females, those with iodine deficiency and people with a history of radiation exposure. Some thyroid function parameters (ie, thyroid-stimulating hormone (TSH) and thyroid antibodies) might contribute to the growth and progression of TNs.[7] TNs are also closely related to a greater waist circumference, higher triglyceride levels, homeostasis model assessment of insulin resistance (HOMA-IR) and glycated haemoglobin (HbA1c).[8 9]

Sex hormones trigger the promotion of sexual maturity, development of sexual characteristics and maintenance of sexual function. Over the last decade, their roles in endocrine and metabolic diseases have also been revealed. Many convincing studies[10–13] reported that low total testosterone (T) and sex hormone-binding globulin (SHBG) levels were strongly associated with metabolic syndrome and type 2 diabetes. T and oestradiol ($E_2$) were positively correlated with markers of insulin resistance, fasting glucose[14 15] and measures of adiposity.[16] It was also recently reported that low follicle-stimulating hormone (FSH) levels were associated with pre-diabetes and diabetes in postmenopausal women.[17]

However, the direct relationship between TNs and reproductive hormone levels has never been reported. We performed a population-based observational Survey on Prevalence in East China for Metabolic Diseases and Risk Factors (SPECT-China) in 2014–2015 to investigate the relationship between TNs and FSH, luteinising hormone (LH), $E_2$, total T and SHBG in Chinese men.

## MATERIALS AND METHODS

### Study participants

SPECT-China[18–20] is a population-based cross-sectional survey on the prevalence of metabolic diseases and risk factors in East China; its registration number is ChiCTR-ECS-14005052 (www.chictr.org.cn). We used a stratified and cluster sampling method. The first level of sampling was stratified by urban and rural areas, and the second level was stratified by economic development areas. From February 2014 to December 2015, this study was performed in Shanghai, Zhejiang, Jiangxi, Jiangsu and Anhui Province, 22 sites in East China where 99.5% of residents are Han Chinese. Adults aged 18 years old and above who were Chinese citizens and lived in their current residence for more than 6 months were invited into our study. Those patients with severe communication problems or acute illness and who were unwilling to participate were excluded. All participants provided written informed consent before data collection. The study protocol was approved by the Ethics Committee of Shanghai Ninth People's Hospital, Shanghai JiaoTong University School of Medicine. All procedures were followed in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1975, as revised in 2008.

There were 4307 men over 18 years of age who were not using hormone replacement therapy. Men with missing main lab results, including the total T, $E_2$, LH, FSH and SHBG levels (n=102); missing ultrasound information (n=150); a history of thyroid surgery or thyroid disease (including hyperthyroidism, hypothyroidism, subacute thyroiditis and prior radioactive iodine treatment) (n=31) were excluded. Finally, 4024 subjects were included in the final analysis. The participants' inclusion and exclusion in this analysis are shown in figure 1.

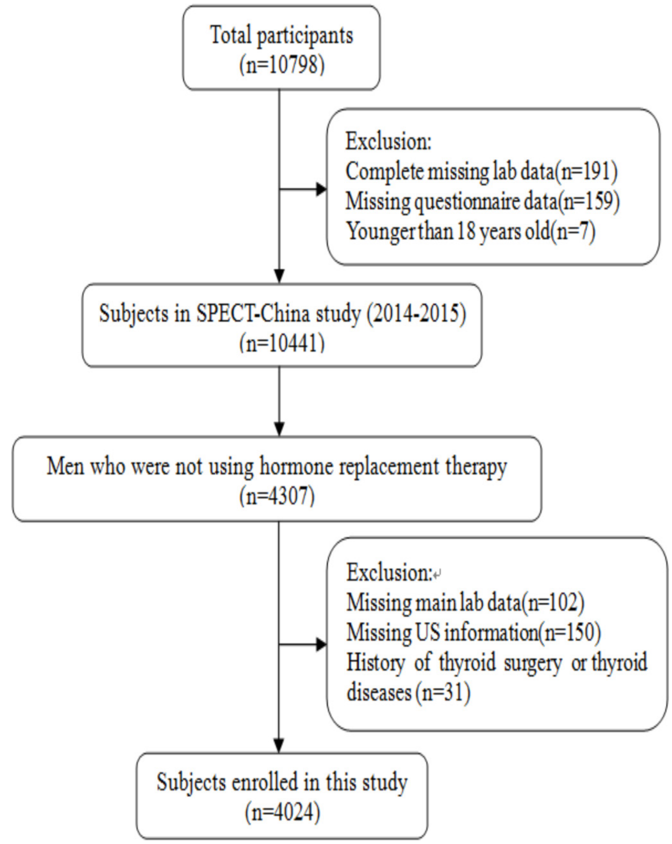


**Figure 1** Flowchart of the participants in this study selected from Survey on Prevalence in East China for Metabolic Diseases and Risk Factors (SPECT-China)

### Data collection

In every step of this study, all data collection were performed by the same staff group from the Department of Endocrinology and metabolism in Shanghai Ninth People's Hospital, Shanghai JiaoTong University School of Medicine. All staff successfully completed a standard training programme that made them familiar with the specific tools and methods used. A standard questionnaire was administered by trained staff to obtain information on the demographic characteristics, personal and family medical history and risk factors in daily life. Clinical staff members were trained to measure blood pressure and obtain anthropometric measurements and blood samples.

### Thyroid US

Thyroid examination was performed by the same two registered US doctors who had a professional certificate for US measurement awarded by the Ministry of Health of China using B-mode US imaging (M7, Mindray ShenZhen, P.R. China).with a 10 MHz linear array probe. The scanning protocol in all cases included both transverse and longitudinal real-time imaging of the thyroid. The characteristics of the thyroid parenchyma were described according to their echogenicity and homogeneity. Nodule characteristics, including solitary or multiple, diameter exceeding 10 mm, microcalcification and a 'taller' than 'wider' shape nodule, were recorded.

### Laboratory assays

Serum samples for laboratory assays were obtained by venepuncture after an 8 hour fast between 07:00 and

10:00 hours in the morning. Blood samples were stored at −20°C when they were collected and were then shipped by air in dry ice to one central laboratory within 2–4 hours of collection, which was certified by the College of American Pathologists.

The total T, $E_2$, FSH and LH levels were measured by chemiluminescence (Siemens, immulite 2000, Erlangen, Germany). SHBG was measured by chemiluminescence immunoassay (Roche Cobas E601, Basel,Switzerland). The minimal detectable limit for each sex hormone was as follows: 0.7 nmol/L (total T), 73.4 pmol/L ($E_2$), 0.1 IU/L (FSH and LH) and 0.35 nmol/L (SHBG). The interassay coefficients of variation were as follows: 6.6% (total T), 7.5% ($E_2$), 4.5% (FSH), 6.0% (LH) and 7% (SHBG). The intra-assay coefficients of variation were as follows: 5.7% (total T), 6.2% ($E_2$), 3.8% (FSH), 4.9% (LH) and 7% (SHBG). HbA1c was assessed by high-performance liquid chromatography (MQ-2000PT, Medconn, Shanghai, China). Plasma glucose and lipid profiles, including the total cholesterol, triglycerides, high-density lipoprotein (HDL) and low-density lipoprotein (LDL), were measured by Beckman Coulter AU680 (Brea, USA). Insulin was detected using the chemiluminescence method (Abbott i2000 SR,Chicago , USA). The serum antithyroid peroxidase antibody (TPOAb), thyroglobulin antibody (TgAb), thyroxine ($T_4$), triiodothyronine ($T_3$), and TSH levels were measured using a chemiluminescence immunoassay (Siemens, immulite 2000, Erlange, Germany).

### Definition

Nodule: ≥2 mm in diameter

Multiple nodules: ≥2 nodules and ≥2 mm in diameter, in one or both lobes

Microcalcification: calcification <2 mm

TPOAb and TgAb positive: >60 U/mL

### Statistical analysis

We performed survey analyses with IBM SPSS Statistics V.22(IBM Corporation, Armonk, NY, USA). All analyses were two sided. A p value <0.05 was considered significant. Continuous variables were expressed as the mean (±SD) values and categorical variables are presented as numbers (percentage). Continuous variables were compared using Student's t-test. The Mann-Whitney U test was used for non-normally distributed continuous variables, and the Pearson $\chi^2$ test was used for dichotomous variables. The OR and 95% CIs were calculated using logistic regression to determine the risk of TN for each quartile of SHBG by using the highest quartile as the reference. The base model (model 1) included terms for age and smoking. Model 2 included the terms for model 1 and the body mass index (BMI) and waist-to-hip ratio. Model 3 included the terms for model 2 and the HOMA-IR, HDL, LDL, TG, systolic blood pressure, $T_4$ and TPOAb. Model 4 included the terms for model 3 and the total T. The BMI was calculated as the weight in kilograms divided by the height, in metres, squared. Insulin resistance was estimated by the HOMA-IR index: (fasting insulin (mIU/L))×(FPG (mmol/L))/22.5.

## RESULTS

### Clinical characteristics of participants with and without thyroid nodules

A total of 4024 subjects were enrolled in this study. The mean age was 54.15±13.08 years and the mean BMI was 24.86±3.33 kg/m$^2$. Among them, 1667 participants had TNs. The prevalence of TN was 41.4%. Compared with those without TNs (TN(−)), men with TNs (TN(+)) were significantly older and had a significantly greater BMI, waist-to-hip ratio and systolic pressure. These men also had higher levels of HOMA-IR, HbA1c, LDL, TG, $T_4$, TPOAb and TgAb and lower levels of HDL (p<0.05). The percentages of TPOAb positive and TgAb positivity were higher in the TN(+) group, but there was no significant difference (p>0.05) (table 1).

### Analysis of covariance in age subgroups

To control for the influence of age, further analysis of covariance in age subgroups was performed (table 2). In the TN(+) group, both the total T and SHBG levels were significantly lower than in the TN(−) group in both age subgroups (p<0.05). The $E_2$/T ratio level was significantly higher in the TN(+) groups in both age subgroups (p<0.05). FSH was significantly higher in the TN(+) group in men under 60 years of age. No significant difference for $E_2$ or LH was detected in any age subgroup.

### Prevalence of TN according to sex-related hormone levels

To more effectively determine the relationship between the sex-related hormone levels and TN, we analysed the association between the serum sex hormone levels and the prevalence of TN. We classified subjects according to sex hormone level quartiles and age. We observed that the prevalence of TN decreased with increasing SHBG levels (56.5%, 54%, 51.9% and 46.2% in men over 60 years of age and 39%, 35.5%, 34.8% and 30.4% in men under 60 years of age; p<0.05 in both age subgroups). The prevalence of TN decreased with increasing SHBG levels. For the total T, although there was a significant difference, the prevalence of TNs was not continuous with the total T level. No significant differences for $E_2$, $E_2$/T and LH were detected in any age subgroups (table 3).

### Multivariable analyses

Given the findings of lower SHBG levels in subjects in the TN(+) group and the prevalence of TN increasing with decreasing SHBG levels, we evaluated the adjusted ORs for men in the TN(+) group. Table 4 shows the ORs according to SHBG quartiles using binary logistic regression analyses. Among men, decreasing quartiles of SHBG were positively associated with TNs (p for trend <0.05). Men in the lowest quartile of SHBG had a 1.91-fold risk of developing TNs compared with those in the highest quartile (95% CI 1.54 to 2.37; table 4, model

Table 1 General characteristic of all subjects with and without thyroid nodules (TNs)

| | All | TN(–) | TN(+) | p Value |
|---|---|---|---|---|
| N (%) | 4024 (100) | 2357 (58.6) | 1667 (41.4) | – |
| Age (years)* | 54.15±13.08 | 51.96±13.13 | 57.26±12.35 | <0.001 |
| Smokers (n(%)) | 2098 (52.1) | 1208 (51.3) | 890 (53.3) | 0.135 |
| Metabolic factors | | | | |
| BMI (kg/m$^2$)* | 24.86±3.33 | 24.65±3.40 | 25.17±3.22 | <0.001 |
| Waist-to-hip ratio* | 0.89±0.07 | 0.89±0.07 | 0.90±0.08 | <0.001 |
| HOMA-IR | 1.49±2.80 | 1.46±2.94 | 1.54±2.57 | 0.016 |
| HbA1c (%) | 5.62±1.02 | 5.54±0.94 | 5.73±1.11 | <0.001 |
| LDL (mmol/L)* | 3.08±0.76 | 3.05±0.76 | 3.13±0.75 | 0.003 |
| HDL (mmol/L)* | 1.36±0.32 | 1.37±0.32 | 1.33±0.32 | <0.001 |
| TG (mmol/L) | 1.89±1.91 | 1.88±2.02 | 1.90±1.73 | 0.021 |
| TC (mmol/L)* | 5.14±1.04 | 5.13±1.03 | 5.16±1.05 | 0.321 |
| Systolic pressure (mm Hg)* | 134.58±20.81 | 133.43±20.65 | 136.22±20.92 | <0.001 |
| Thyroid hormones | | | | |
| TSH (mIU/L) | 2.46±3.20 | 2.44±2.70 | 2.47±3.78 | 0.570 |
| $T_3$ (nmol/L) | 1.79±0.48 | 1.79±0.51 | 1.79±0.44 | 0.647 |
| $T_4$ (nmol/L)* | 113.88±22.72 | 112.95±22.48 | 115.18±23.00 | 0.002 |
| TPOAb (U/ml) | 69.36±204.21 | 65.46±196.92 | 74.86±214.02 | <0.001 |
| TPOAb (+) (n(%)) | 332 (8.3) | 179 (7.6) | 153 (9.2) | 0.073 |
| TgAb (+) (U/ml) | 27.97±61.62 | 26.24±54.47 | 30.41±70.44 | 0.001 |
| TgAb (+) (n(%)) | 232 (5.8) | 131 (5.6) | 101 (6.1) | 0.506 |

Data were summarised as the mean ±SD for continuous variables or as a number with proportion for categorical variables.
*These data were normal distribution.
BMI, body mass index; HbA1c, glycated haemoglobin; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; $T_3$, triiodothyronine; $T_4$, thyroxine; TC, total cholesterol; TG, triglycerides; TgAb, thyroglobulin antibody; TN(+), participants with thyroid nodule(s); TN(–), participants without thyroid nodule; TPOAb, thyroid peroxidase antibody; TSH, thyroid-stimulating hormone; Smokers: smoking history including current and past.

1). This association remained statistically significant after fully adjusting for age, smoking history (including current and past), BMI, waist-to-hip ratio, HOMA-IR, HDL, LDL, TG, systolic blood pressure, $T_4$, TPOAb and total T (p for trend=0.017). For the lowest compared with the highest SHBG quartile, the OR of the TN was 1.42 (95%CI 1.07 to 1.89; table 4, model 4).

### Association between thyroid nodule features and sex-related hormones

According to the quartiles of sex-related hormone levels, we calculated the percentage of each sonographic feature in the TN(+) group (n=1667), particularly for US characteristics associated with malignancy (nodule >10 mm, microcalcification and a 'taller' than 'wider' shape).[6]

Table 2 Sex-related hormones of all subjects with and without thyroid nodules (TNs)

| | ≥60 years | | | <60 years | | |
|---|---|---|---|---|---|---|
| | TN(–) | TN(+) | p | TN(–) | TN(+) | p |
| N(%) | 724 (47.9) | 788 (52.1) | – | 1632 (65.0) | 879 (35.0) | – |
| Total T (nmol/L) | 18.73±7.31 | 17.66±6.51 | 0.006 | 16.19±5.48 | 15.36±5.25 | 0.001 |
| $E_2$ (pmol/L) | 128.17±71.55 | 128.14±64.91 | 0.791 | 98.36±55.44 | 97.10±54.04 | 0.618 |
| $E_2$/T ratio | 7.58±6.12 | 7.91±4.64 | 0.023 | 6.54±5.32 | 7.56±19.16 | 0.033 |
| FSH (IU/L) | 12.57±9.96 | 12.72±10.67 | 0.893 | 6.67±4.60 | 7.10±4.67 | 0.002 |
| LH (IU/L) | 7.56±4.83 | 7.61±5.03 | 0.916 | 4.76±2.69 | 4.89±2.48 | 0.085 |
| SHBG (nmol/L) | 63.64±29.58 | 58.71±26.25 | 0.002 | 39.33±20.17 | 36.58±16.69 | 0.006 |

$E_2$, oestradiol; FSH, follicle-stimulating hormone; LH, luteinising hormone; SHBG, sex hormone-binding globulin; T, testosterone; TN(+); participants with thyroid nodule(s); TN(-); participants without thyroid nodule.

**Table 3** The prevalence of thyroid nodules (TNs) by quartiles of sex related hormone levels

| | ≥60 years | | | | | <60 years | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Q1 | Q2 | Q3 | Q4 | p | Q1 | Q2 | Q3 | Q4 | p |
| Total T (nmol/L) | <13.20 | 13.20–17.13 | 17.13–22.00 | >22.00 | – | <12.30 | 12.30–15.20 | 15.20–18.70 | >18.70 | – |
| TN(+)(n(%)) | 219 (57.2) | 191 (51.2) | 202 (53.2) | 176 (46.8) | 0.037 | 235 (37.0) | 238 (37.4) | 229 (36.9) | 177 (28.6) | 0.002 |
| $E_2$ (pmol/L) | <87.75 | 87.75–119.00 | 119.00–162.00 | >162.00 | – | <41.50 | 41.50–94.90 | 94.90–126.00 | >126.00 | – |
| TN(+)(n(%)) | 192 (50.8) | 201 (51.7) | 198 (53.8) | 197 (52.3) | 0.869 | 222 (35.4) | 226 (35.7) | 219 (34.7) | 212 (34.2) | 0.951 |
| $E_2$/T ratio | <4.46 | 4.46–6.70 | 6.70–9.64 | >9.64 | – | <3.67 | 3.67–5.96 | 5.96–8.29 | >8.29 | – |
| TN(+)(n(%)) | 184 (48.7) | 192 (50.8) | 205 (54.2) | 207 (54.8) | 0.284 | 211 (33.9) | 198 (31.3) | 234 (37.3) | 236 (37.6) | 0.062 |
| FSH (IU/L) | <7.20 | 7.20–9.80 | 9.80–14.30 | >14.30 | – | <4.10 | 4.10–5.80 | 5.80–8.11 | >8.11 | – |
| TN(+)(n(%)) | 197 (51.2) | 195 (52.1) | 203 (53.6) | 193 (51.6) | 0.782 | 199 (31.5) | 215 (33.2) | 218 (36.1) | 247 (39.4) | 0.018 |
| LH (IU/L) | <4.70 | 4.70–6.50 | 6.50–9.06 | >9.06 | – | <3.10 | 3.10–4.22 | 4.22–5.90 | >5.90 | – |
| TN(+)(n(%)) | 201 (51.7) | 198 (52.7) | 199 (53.9) | 190 (50.3) | 0.782 | 217 (33.1) | 211 (35.2) | 218 (33.9) | 233 (38.1) | 0.267 |
| SHBG (nmol/L) | <40.30 | 40.30–56.10 | 56.10–75.90 | >75.90 | – | <24.90 | 24.90–34.50 | 34.50–47.30 | >47.30 | – |
| TN(+)(n(%)) | 214 (56.5) | 204 (54.0) | 196 (51.9) | 174 (46.2) | 0.032 | 245 (39.0) | 226 (35.8) | 218 (34.8) | 190 (30.4) | 0.015 |

$E_2$, oestradiol; FSH, follicle-stimulating hormone; LH, luteinising hormone; SHBG, sex hormone-binding globulin; T, testosterone; TN(+); participants with thyroid nodule(s).

Only the interquartile comparison of $E_2$ with a 'taller' than 'wider' shape and LH with multiple nodules was significantly different in males who over 60 years of age ($p<0.05$). However, the trend of these percentages fluctuated (online supplementary table 1).

We also evaluated the association of sex-related hormones and US characteristics associated with malignancy (nodule >10 mm, microcalcification and a 'taller' than 'wider' shape) using binary logistic regression analyses. After fully adjusting for age, smoking history, BMI, TSH and TPOAb, there was no significant association between sex-related hormones and these three US characteristics associated with malignancy ($p>0.05$).

## DISCUSSION

A previous study reported that TNs are more common in elderly people and had a significantly higher frequency of thyroid antibody positivity, obesity, insulin resistance and diabetes mellitus.[6–9] In agreement with prior reports, men with TNs in our study were older and had higher TPOAb and TgAb levels; greater BMI and waist-to-hip circumference and higher triglyceride, LDL, HOMA-IR and HbA1c levels.

As we previously described, TNs were associated with glucose and lipid metabolism, whereas the latter was closely related to sex hormones. Little was known about the association between TNs and sex hormone levels. Therefore, we performed a full adjustment, including for age, BMI, waist-to-hip ratio, glucose and lipid metabolism factors, thyroid parameters and total T, which might contribute to the growth and the progression of TNs and be related to sex hormones, to investigate the issue above. In this population-based study, the association remained significant after full adjustment. Lower quartiles of SHBG had greater risks of TNs. The fully adjusted OR of TNs increased by 42% for the lowest quartile compared with highest quartile of SHBG levels, which confirmed that lower SHBG levels were significantly associated with TN among men.

SHBG is the main transport binding protein for sex steroid hormones in plasma, and it regulates their accessibility to target cells. The human liver secretes SHBG into the blood, where it binds androgens and oestrogens with high affinity, regulating their bioavailability. The plasma SHBG level has been reported as a biomarker of several endocrine and metabolism diseases, including obesity, thyroid hormone disorders, polycystic ovary syndrome, Cushing's syndrome and acromegaly.[21] Epidemiological studies have shown that the SHBG levels are predictive of a higher risk for developing metabolic syndrome[12 22] and type 2 diabetes, which was explained by insulin resistance.[23 24] SHBG is positively associated with HDL and inversely associated with BMI[12] and triglycerides.[13] Thyroid hormones influence the plasma SHBG levels under normal and pathological conditions by altering hepatic SHBG production.[21] To the best of our knowledge, this study is the first to detect the association between TNs

Table 4 Association of sex hormone-binding globulin with thyroid nodule

| | Model 1 | Model 2 | Model 3 | Model 4 |
|---|---|---|---|---|
| Q1 (<28.70) | 1.91 (1.54, 2.37) | 1.62 (1.29, 2.04) | 1.61 (1.26, 2.06) | 1.42 (1.07, 1.89) |
| Q2 (28.70–41.20) | 1.52 (1.25, 1.86) | 1.38 (1.12, 1.70) | 1.34 (1.07, 1.67) | 1.21 (0.95, 1.56) |
| Q3 (41.20–59.30) | 1.34 (1.10, 1.62) | 1.25 (1.03, 1.52) | 1.22 (1.00, 1.49) | 1.14 (0.92, 1.42) |
| Q4 (>59.30) | 1.00 | 1.00 | 1.00 | 1.00 |
| p for trend | <0.001 | <0.001 | <0.001 | 0.017 |

The data are expressed as OR (95 % CI) unless otherwise indicated. Logistic regression analysis was used.
Model 1 included terms for age, smoking.
Model 2 included terms for model 1, BMI, waist-to-hip ratio.
Model 3 included terms for model 2, HOMA-IR, HDL, LDL, TG, and systolic blood pressure, $T_4$ and TPOAb.
Model 4 included terms for model 3, total T.
BMI, body mass index; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; LDL, low-density lipoprotein; T, testosterone; $T_4$, thyroxine; TG, triglycerides; TPOAb, thyroid peroxidase antibody.

and sex hormones in such a large population; the association was independent of age, BMI, waist-to-hip ratio, metabolic and lipid factors, thyroid parameters and total T.

Low levels of SHBG have been consistently documented in low-grade chronic inflammatory diseases in which there are changes in the proinflammatory or anti-inflammatory cytokines, such as TNF-α, IL-1β and adiponectin.[21 25] Several studies have demonstrated that there was a negative correlation between SHBG and leptin.[26 27] Leptin has some impact on the thyroid dysfunction through disrupting the feedback loop of the HPT axis by altering TRH[28] and thyroid autoimmunity[29] via inflammation and inflammatory molecules (primarily TNF-α).[30] Leptin expression was also strongly correlated with the thyroid volume,[31] a larger tumour size[32] and thyroid cancer subtypes.[33] We hypothesised that a lower SHBG level might play a role in the pathogenesis of TNs by changing the leptin level.

In our survey, we calculated the percentage of each sonographic feature in the TN(+) group, especially for US characteristics associated with malignancy (nodule >10 mm, microcalcification and a 'taller' than 'wider' shape)[6] and tried to analyse the association between sex-related hormones and thyroid US characteristics associated with malignancy. However, no significant difference was found. The relatively small percentage of participants with malignant US characteristics and the limitations of ultrasound diagnosis of malignancy may affect the results.

Our study has several strengths. First, we evaluated a relatively large sample of men to examine the relationship between sex hormones and TNs, which have not been reported before. Second, anthropometric measurements and questionnaires were completed by the same trained research group, and all thyroid ultrasound examinations were performed by the same two doctors with strong quality control. Third, community-dwelling subjects living in multiple sites in China were recruited such that the results may be more representative compared with a clinic-based population.

However, our study also has several limitations. First, owing to the cross-sectional study nature, no causal inference can be drawn, and a reverse effect of TN leading to changes in SHBG still needs to be excluded. Prospective studies are needed to clarify the precise interrelationship. Second, we only measured sex hormones a single time to characterise each man's hormonal status. However, this may not have significantly affected the results because a single measurement on morning samples could provide representative and reliable data in large epidemiological studies.[34] Third, the gold standard for diagnosing a TN is biopsy, and the use of thyroid US has limitations. However, thyroid biopsy may not be feasible in such a large sample. Fourth, although TNs are more common in females,[6] female subjects were not enrolled in this study for many reasons. The values of total T measured by the IMMULITE 2000 platform are not reliable when they are at low concentrations, as observed in women.[35] The phases of the menstrual cycle and age of menopause for the women were not available. In addition, the effect of pregnancy on TN formation was considered.[36] For these reasons, we could not make an adequate assessment of the association between TNs and sex-related hormones in women.

## CONCLUSION

TNs are highly prevalent in males in China. A lower SHBG level was significantly associated with TN among men. The potential role of SHBG in the pathogenesis of the TN remains to be elucidated.

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