Environmental Paraben Exposure and Risk of Thyroid Cancer and Benign Nodules: A Pilot Study

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Abstract

Parabens are widely used as preservatives, which have been found to affect thyroid function in toxicological studies. However, population studies on whether they are associated with thyroid tumor remain unclear. This study aims to investigate the relationship between environmental paraben exposure and thyroid cancer and benign nodules. The detectable percentages of methyl paraben, ethyl paraben, and propyl paraben in the urinary samples of 425 study subjects were 99.06%, 95.29% and 92.00%, respectively. In the single pollutant model, we found statistically significant difference between certain parabens and thyroid cancer/ benign nodules. Further, we found the mixture effect of parabens on increased risk of thyroid cancer (OR = 0.24, 95% CI: 0.18, 0.31) and benign nodule (OR =1.33, 95% CI: 0.86, 1.80). The significant gender-associated effects were found in associations with certain parabens and thyroid cancer and benign nodules (Both P for interactions < 0.05). Overall, our results showed that individual exposure of paraben mixtures may be associated with the risk of thyroid cancer and benign nodules, and there were gender differences.

1. Introduction

Thyroid cancer is a common endocrine malignancy, accounting for less than 2% of all types of tumors, but it has been the fourth most common cancer worldwide [1]. Thyroid cancer incidence, especially papillary thyroid carcinoma, has shown a steadily increasing trend globally [2–4]. The substantial increasing rates of both thyroid cancer incidence and incidence-related mortality have been well-documented in the United States from 1974 to 2013 [5]. Moreover, a recent study has reported the prevalence of thyroid nodule in Chinese population [6]. Though overall 5-year relative survival for thyroid cancer is quite high (> 90% for men and women) [7], the increasing number of survivors need to face social, financial, and spiritual burden over their lifetimes due to the continued surveillance and prolonged treatment.

Changes in detection and diagnostic procedures, such as improved imaging techniques, can only explain approximately 50% of the rise in thyroid cancer over the past 20 years [8]. Another part of the identified and possible risk factors for thyroid cancer are environmental pollutant exposure, excessive and deficient iodine intake, obesity, etc. [5]. Recently, emerging evidences have documented that certain endocrine-disrupter chemical may also be responsible for the increasing incidence of thyroid tumor [9–11]. Thus, there is an urgent need to better understand the effect of environmental endocrine-disruptors on thyroid disease.

Parabens are used extensively as preservatives in foods, beverages, cosmetics and pharmaceutical products [12–14]. Humans can be exposed to parabens predominantly through ingestion of paraben-containing drugs and foodstuff [13, 15]. After intake, parabens are readily metabolized and are rapidly excreted into the urine. Urinary paraben has been established as a valid biomarker for human biomonitoring to characterize exposure levels [16–18]. In particular, high detection frequencies (≥ 98%)

of several parabens including methyl-paraben (MeP), ethyl-paraben (EtP), and propyl-paraben (PrP) have been observed in urine samples [19, 20].

Experimental studies have shown that parabens can exhibit endocrine-disrupting properties [21]. For example, given their structural similarity, parabens are likely to exert thyroid disrupting effects [22]. Parabens have thyrotoxic effects in the female Sprague-Dawley rat model, where they are associated with thyroxine (T4) levels and cause thyroid weight loss [10]. And, T4 are associated with cancer progression, including thyroid tumor [23, 24]. Moreover, changes in thyroid hormone levels such as triiodothyronine (T3) and T4 in the blood affect thyroid stimulating hormone (TSH) changes, resulting in bilateral thyroid follicular cell hypertrophy and increased mitotic activity in Wistar rat model [25]. Indeed, altered levels of TSH can lead to tumor growth and thyroid cancer initiation in mice with a thyroid-specific knock-in of oncogenic Braf [26, 27]. Also, abnormally elevated TSH can increase the risk of developing thyroid cancer [28]. Mutations in TSH receptors may occur, causing pathological changes in thyroid function in response to altered hormone signaling, stimulating thyroid cells to secrete and release large amounts of bioactive factors such as cell growth factors and vascular endothelial growth factors, which in turn lead to tumorigenesis [29].

Epidemiological studies have linked the exposure to a suite of parabens with thyroid-function disruption in human, such as the disrupted homeostasis of serum THs and TSH [30–33]. A pregnancy study reported that parabens are associated with thyroid hormone levels such as free thyroxine (FT4), T3, and T4 [34]. Baker et al. observed that urine MeP associated with TSH, free T4, and total T3 [20]. Moeller et al. found that, altered TH levels may increase the incidence of multiple tumors [27, 35, 36]. Abnormal TSH levels have also been related to increased risk of thyroid cancer from an US study [37, 38].

To data, very few studies examined the associations between paraben exposure and thyroid cancer and benign nodules. To address a key data gap, a case-control study was conducted in Chinese population to investigate the relationship of paraben exposure with thyroid cancer and benign nodules, and improve our understanding of the risk of thyroid tumor caused by exposure to parabens.

2. Materials And Methods

2.1 Study population.

Participants were recruited from March to December 2016 at the Department of Thyroid and Breast Surgery at Wuhan Central Hospital, Wuhan, China. We restricted the recruitment to study subjects who met the following criteria: endocrine disease (e.g., chronic renal disease and hypertension) and occupational exposure (e.g., lacquerware, pesticides, polyvinyl chloride, and industrial solvents). As such, patients diagnosed with thyroid disease requiring further surgery were recruited, and further divided into two groups based on postoperative pathology reports (benign nodule and thyroid cancer): thyroid cancer group (144 patients) and benign nodule group (137 patients). In addition, we recruited 144 healthy participants who sought health screening at the same hospital as controls. Due to gender differences in

the incidence rate of thyroid diseases, the thyroid cancer and benign nodule groups were gender-matched to the control group. The Ethics Committee of Jinan University approved our study protocol.

2.2 Questionnaire and sample collection.

After obtaining the participants' informed consent, each participant was subjected to completing a questionnaire about demographic characteristics, lifestyle factors, dietary habits, and medical history. In the case of female participants, we collected additional information regarding their reproductive history (e.g., menstrual status and menopausal status). We defined current smokers as individuals who reported smoking ≥ 100 cigarettes throughout their lifetime, whereas former smokers were respondents who reported smoking ≥ 100 cigarettes during their lifetime but currently quit smoking. The "current drinkers" were defined as individuals who consumed alcohol more than once a week. Prior to any medication or surgery, all subjects were asked to provide one spot first morning urine samples. All samples were stored at -80°C until laboratory determinations.

2.3 Exposure estimate.

We determined urinary MeP, EtP, and PrP using previously described method with minor modifications [39]. In brief, 0.5 mL of urine were thawed at room temperature and transferred into 15 mL polypropylene tubes, followed by adding 10 μL of β-glucuronidase (≥ 100000 units/mL, from Helix pomatia, Sigma-Aldrich) and 200 μL ammonium acetate buffer (pH = 5.0). The mixtures were incubated overnight at 37°C. Then, the samples were spiked with 10 μL of an internal standard (tert-butyl paraben-d9), and subsequently extracted three times in a methyl tert-butyl ether/ethyl acetate (5:1; v/v) solution. For optimal extraction, the samples were shaken for 30 min and then centrifuged at 4500 rpm for 10 min. Final extracts were concentrated to near dryness, and reconstituted with 100 μL of the solvent mixture (acetonitrile: water = 6:4) before instrumental analysis.

The target chemicals were determined by an ultraperformance liquid chromatography (UPLC) coupled with an AB Sciex 5500 triple quadrupole mass spectrometer (MS/MS, Toronto, Canada). Chromatographic separation was achieved using an Agilent ZORBAX Extend-C18 column (Narrow Bore RR 2.1 mm×100 mm, 3.5-Micron, 80 Å). The mass spectrometer was equipped with an electrospray ionization (ESI) probe in the negative mode and operated in the multiple reaction monitoring (MRM) mode.

2.4 Quality Control.

Each batch of 20 samples was processed together with a blank control to assess procedural contamination. Final concentrations were corrected for average blank values and recoveries of internal standards. The limits of detection (LODs) for urinary MeP, EtP, and PrP were 0.09, 0.07, and 0.07 μg/L, and the average recoveries for the three targets were 101.7%, 83.7–109.9%, respectively. Measurements below the level of detection (LOD) were replaced with LOD/ $\sqrt{2}$ for further data analysis [40, 41]. Urine dilution was corrected by urinary creatinine based on the Jaffe reaction [40].

2.5 Statistical analysis

2.5.1 Analyses for each paraben.

The demographic characteristics of our study participants were presented as n (%) for categorical variables and mean ± standard deviation (SD) for continuous variables. Differences in demographic characteristics and urinary parabens between cases and the controls were compared using the Chi-square test for categorical variables and the Mann-Whitney U-test for continuous variables. We also analyzed correlations between individual urinary paraben concentrations using the Spearman's rank test.

We applied unconditional logistic regression models to assess the association of creatinine-corrected urinary MeP, EtP and PrP with the risk of thyroid cancer and benign nodule, and calculated odds ratios (ORs) and 95% confidence intervals (CIs). Based on the median urinary creatinine-corrected MeP, EtP and PrP concentrations in the control group, exposure variables were categorized into low-exposure and high-exposure groups. Also, natural logarithm (ln)-transformed urinary MeP, EtP and PrP concentrations were included in the same model as continuous variables. Covariates were included whether exposure changed the risk of thyroid cancer or benign nodule (ORs) > 10%. The body mass index (BMI) and age (continuous), alcohol consumption (yes vs. no), gender (male vs. female), household income (< 5000, 5000-10000, and ≥ 10000 yuan/month), and smoking status (ever and current vs. never) were entered into final model.

Because of potential gender effect in the incidence of thyroid cancer, we further analyzed gender associated differences in urinary creatinine-corrected MeP, EtP and PrP concentrations and the risk of thyroid cancer and benign nodule. The interaction term between parabens and gender was included in the regression model. Statistical analyses were performed using SPSS version 26 (IBM Corporation, NJ, USA). We considered P-value < 0.05 as statistically significant.

2.5.2 Mixture analyses.

We used quantile g-computation to estimate the overall mixture effect of the three parabens on the risk of thyroid cancer and benign nodule [42]. The quantile g-computation based implementation of g-computation can yield consistently estimates of the effect of increasing all exposures by one quantile, simultaneously. Each paraben was defined as a weight by fitting a generalized linear model between covariates, exposures, and outcomes. The weights can represent the contribution of each paraben to the overall mixture effect. The sum of regression coefficients across multiple paraben exposures can be estimated by quantile g-computation. We Mixtures analyses were conducted via the "*gqcomp*" R package in R version 4.0.4.

3. Results

The demographic characteristics of our study participants are presented in Table 1. Most of the participants (75%) never smoked or drank alcohol. The significant differences were observed in educational background, postmenopause, household income, and age at first pregnant between the benign nodule group and thyroid cancer group compared with the control group, respectively (All P-values

≤ 0.01). The significant differences were additionally observed in age between controls and the benign nodule participants (P-value < 0.01).

Table 1
Distribution of characteristics among study participants a (N = 425).

Characteristics	Control N=144	Benign nodule N=137	P-values	Thyroid cancer N=144	P-values
Age (years)	44.82±10.37	54.26±11.55	< 0.01	47.08±11.60	0.08
BMI (kg/m^2)	22.81±3.03	23.53±4.13	0.10	23.33±3.10	0.16
Gender			0.99		0.90
Male	41 (28.47)	39 (28.47)		40 (27.78)	
Female	103 (71.53)	98 (71.53)		104 (72.22)	
Education			< 0.01		< 0.01
Less than high school	18 (12.77)	42 (31.34)		33 (23.08)	
High school	19 (13.48)	59 (44.03)		50 (34.97)	
College and above	104 (73.75)	33 (24.63)		60 (41.95)	
Household income			< 0.01		< 0.01
<5000	40 (28.17)	74 (56.06)		59 (43.70)	
5000-10000	41 (28.87)	46 (34.85)		57 (42.22)	
≥10000	61 (42.96)	12 (9.09)		19 (14.08)	
Smoking status			0.46		0.87
Never	120 (83.33)	99 (79.84)		111 (84.09)	
Former & current	24 (16.67)	25 (20.16)		21 (15.91)	
Alcohol use			0.06		0.18
No	120 (84.51)	94 (75.20)		104 (78.20)	
Yes	22 (15.49)	31 (24.80)		29 (21.80)	
Age of menarche (years)	13.16±1.64	13.90±1.97	< 0.01	13.56±1.82	0.10
Postmenopause			< 0.01		< 0.01
No	37 (37.00)	69 (69.70)		60 (57.14)	
Yes	63 (63.00)	30 (30.30)		45 (42.86)	
Age at menopause (years)	49.09±5.84	49.46±3.73	0.70	49.14±4.06	0.96

Characteristics	Control N=144	Benign nodule N=137	P-values	Thyroid cancer N=144	P-values
Age at first pregnant (years)	25.92±2.86	24.58±3.12	< 0.01	24.85±2.78	0.01
Parity			0.68		0.70
0	10 (9.90)	9 (8.82)		10 (9.26)	
1	75 (74.26)	72 (70.59)		85 (78.70)	
2 and above	16 (15.84)	21 (20.59)		13 (12.04)	

[a] The data were listed as mean±SD for continuous variables and n (%) for categorical variables. P-values were calculated by appropriate parametric or nonparametric methods.

Table 2 presents the distributions of urinary paraben concentrations. The detectable percentages in urinary samples for MeP, EtP, and PrP were 99.06%, 95.29%, and 92.00%, respectively. The urinary parabens concentration ranged from 0.39 to 48.73μg/L. The median concentrations of urinary MeP in group of benign nodule and group of thyroid cancer were higher than those in control. The median concentrations of creatinine-corrected urinary parabens among female population were higher than those in male population. All uncorrected and creatinine-corrected parabens were moderately correlated with one another (Spearman r values ranging from 0.30 to 0.66) (see Figure 1).

Table 2
The distributions of urinary paraben concentrations in study population (N = 425).

Parabens	% > LOD	Median (95% CI)				
		Control	Benign nodule	Thyroid cancer	Female	Male
Unadjusted (μg/L)						
MeP	99.06	29.30 (17.08, 38.37)	48.73 (34.17, 61.73) *	38.05 (27.66, 50.08)	44.26 (37.35, 51.28)	11.62 (8.91, 21.27) **
EtP	95.29	0.54 (0.37, 0.79)	6.07 (3.36, 13.33) *	9.89 (5.58, 18.82) *	4.25 (2.65, 6.40)	2.68 (0.71, 5.14)
PrP	92.00	0.99 (0.59, 2.94)	3.14 (1.82, 7.03) *	2.62 (1.53, 4.18)	3.70 (2.39, 5.51)	0.60 (0.34, 0.93) **
Creatinine-adjusted (μg/g)						
MeP	-	19.18 (12.07, 26.22)	43.52 (34.87, 54.66) *	44.04 (32.20, 55.74) *	44.46 (38.78, 52.28)	8.14 (4.36, 16.16) **
EtP	-	0.41 (0.21, 0.63)	9.03 (5.12, 17.64) *	16.84 (7.48, 25.13) *	5.35 (3.16, 9.03)	1.25 (0.81, 5.30) **
PrP	-	0.86 (0.63, 2.12)	4.35 (2.16, 6.60) *	3.35 (2.06, 5.38) *	4.65 (3.03, 6.45)	0.39 (0.24, 0.89) **

Abbreviations: LOD, limit of detection; CI: confidence interval.

*, the differences of urinary parabens in group of benign nodule and group of thyroid cancer comparing with group of control, respectively.

**, the differences of urinary parabens between female group and male group.

Table 3 shows the associations of creatinine-corrected urinary paraben with the risk of thyroid cancer and benign nodule. After adjusting for possible confounders, we found that urinary MeP and EtP were associated with increased risk of benign nodules (OR: 3.48, 95%CI: 1.74-6.96; OR: 9.82, 95%CI: 4.25-22.67, respectively). Moreover, the higher exposure group for MeP and EtP were associated with a higher risk of benign nodules compared with the lower exposure group (Both P for trends < 0.001). All three parabens, i.e., MeP, EtP and PrP, were associated with increased risk of thyroid cancer (OR: 2.49, 95%CI: 1.38-4.50; OR: 10.03, 95%CI: 4.77-21.09; OR: 2.26, 95%CI: 1.27-4.03). The risk of thyroid cancer was higher among subjects in the higher exposure group for EtP (P for trends < 0.001), compared with the lower exposure group. The above-observed associations remained largely unchanged when MeP, EtP and PrP were included as continuous variables.

Table 3
Associations between urinary parabens (μg/g) and the risks of thyroid cancer and benign nodule in study population.

Parabens	Benign nodule		Thyroid cancer	
	OR (95%CI) [a]	P for trend	OR (95%CI) [a]	P for trend
Methyl Paraben		< 0.001		0.002
Low exposure group	1		1	
High exposure group	3.48 (1.74, 6.96)		2.49 (1.38, 4.50)	
Continuous [b]	1.46 (1.19, 1.80)		1.49 (1.24, 1.78)	
P for continuous	< 0.001		< 0.001	
Ethyl Paraben		< 0.001		< 0.001
Low exposure group	1		1	
High exposure group	9.82 (4.25, 22.67)		10.03 (4.77, 21.09)	
Continuous [b]	1.71 (1.44, 2.02)		1.70 (1.48, 1.96)	
P for continuous	< 0.001		< 0.001	
Propyl Paraben		0.06		0.01
Low exposure group	1		1	
High exposure group	1.84 (0.97, 3.51)		2.26 (1.27, 4.03)	
Continuous [b]	1.14 (1.00, 1.30)		1.26 (1.11, 1.43)	
P for continuous	0.05		< 0.001	

[a] The model was adjusted for gender, age, BMI, alcohol use, smoking status, and household income.

[b] Natural logarithm (ln)-transformed value.

The relationships of creatinine-corrected urinary parabens with the risk of thyroid cancer and benign nodule stratified by gender are showed in Tables 4 and 5, respectively. The significant gender-associated effects were found in associations with urinary MeP and EtP and benign nodule (Both P for interactions < 0.05). After stratifying by gender, urinary MeP and EtP above the median showed an increased risk of benign nodule compared with those below the median (OR: 2.90, 95%CI: 1.28-6.56; OR: 6.85 95%CI: 2.57-18.27, respectively) in females. Moreover, significant interaction effect was also observed between urinary EtP exposures and gender on the thyroid cancer (P for interaction = 0.04). After stratifying by gender, urinary EtP levels above the median showed an increased risk of benign cancer compared with those below the median (OR: 5.65, 95%CI: 2.41-13.22, respectively) in females.

Table 4
Adjusted ORs for urinary parabens (μg/g) and thyroid benign nodule stratified by gender.

Parabens	Benign nodule /control		Adjusted OR (95%CI) [a]		P for interaction
	Female	Male	Female	Male	
Methyl Paraben					0.04
Low exposure group	22/36	15/36	ref	ref	
High exposure group	76/67	24/5	2.90 (1.28, 6.56)	10.58 (2.21, 50.70)	
P for trend			0.01	0.003	
Ethyl Paraben					0.003
Low exposure group	9/41	6/31	ref	ref	
High exposure group	89/62	33/10	6.85 (2.57, 18.27)	35.44 (5.20, 241.66)	
P for trend			< 0.001	< 0.001	
Propyl Paraben					0.18
Low exposure group	23/39	16/33	ref	ref	
High exposure group	75/64	23/8	1.47 (0.69, 3.13)	4.96 (1.16, 21.26)	
P for trend			0.32	0.03	

Abbreviations: OR: odds ratio; CI: confidence interval; ref: reference group.

[a] The model was adjusted for age, BMI, household income, alcohol use and smoking status.

Table 5
Adjusted ORs for urinary parabens (μg/g) and thyroid cancer stratified by gender.

Parabens	Thyroid cancer /Control		Adjusted OR (95%CI) [a]		P for interaction
	Female	Male	Female	Male	
Methyl Paraben					0.34
Low exposure group	27/36	21/36	ref	ref	
High exposure group	77/67	19/5	1.99 (1.00, 3.95)	6.97 (1.86, 26.16)	
P for trend			0.05	0.004	
Ethyl Paraben					0.04
Low exposure group	10/41	5/31	ref	ref	
High exposure group	94/62	35/10	5.65 (2.41, 13.22)	30.10 (6.71, 135.01)	
P for trend			< 0.001	< 0.001	
Propyl Paraben					0.25
Low exposure group	26/39	21/33	ref	ref	
High exposure group	78/64	19/8	2.10 (1.05, 4.20)	2.76 (0.92, 8.27)	
P for trend			0.04	0.07	

Abbreviations: OR: odds ratio; CI: confidence interval; ref: reference group.

[a] The model was adjusted for age, BMI, household income, alcohol use and smoking status.

After adjusting for covariates, using quantile g-computation, our data revealed a one quartile increase in urinary paraben mixtures of 0.24 (95% CI: 0.18, 0.31) in thyroid cancer and 1.33 (95% CI: 0.86, 1.80) in benign nodule, respectively (see Figure 2). Within these models, urinary PrP concentrations were weighted negatively in benign nodule (Figure 2a), in comparison, urinary MeP and EtP were both weighted positively in benign nodule. However, the positive weights of MeP and EtP indicated that parabens may be an important driver of the overall positive association between the paraben exposure and benign nodule. With regard to the effect of these parabens on thyroid cancer, our model showed all positive weights of urinary MeP, EtP, and PrP (Figure 2b).

4. Discussion

In our study, we applied urinary biomarkers to explore the associations of exposure to parabens with the risk of thyroid cancer and benign nodules. We found that urinary MeP and EtP were positively associated with increased the risk of thyroid cancer and benign nodules; and urinary PrP was positively associated with the risk of thyroid cancer. The interaction effects between EtP and gender on the benign nodule and thyroid cancer was also observed. In addition, we assessed the mixture effect of exposure to several parabens using quantile g-computation, and found overall positive associations of exposure with benign nodule and/or thyroid cancer.

To the best of our knowledge, exposure to endocrine disrupting chemicals such as phthalate metabolites, flame retardant chemicals is associated with thyroid function and an increased risk of thyroid cancer [43, 44]. The associations of paraben exposures with thyroid cancer and benign nodule have not been investigated prior to our study. Nonetheless, it has been established that parabens may increase the risk of thyroid disease by disrupting endogenous hormones. Epidemiological studies have shown that exposure to individual parabens disrupted homeostasis of serum THs [30–33]. In addition, chronic thyroid stimulation has been linked to an increased prevalence of toxic nodular thyroid and hypothyroidism in the hypothyroid endemic cretinism population in Uele (Congo) [45, 46]. Moreover, higher TSH levels in patients with benign nodules have been associated with higher risks of differentiated thyroid cancer and advanced tumor stages [47]. Serum TSH levels in the abnormal range have also been associated with the risk of developing thyroid cancer [37, 38]. Therefore, exposure to parabens may interfere with the homeostasis of thyroid hormones, stimulate thyroid cells to secrete and release large amounts of bioactive factors such as cell growth factors and vascular endothelial growth factors, which in turn lead to the development of benign nodules and thyroid cancer [29] such as observed in the present study.

The underlying mechanisms remained unclear between paraben exposures and risk of thyroid cancer and benign nodule. In rodent models, paraben exposure directly affected TH levels [48], while elevated TH can lead to thyroid hypertrophy and consequent weight gain [25], as well as stimulate tumor growth and metastasis [27]. Parabens have exhibited estrogenic and androgenic properties due to similar structures [49, 50], which may also explain their effect on TH regulation and homeostasis. Furthermore, TH can activate intracellular signaling pathways during the progression of cancer at concentrations under the hypothyroidism or thyroid state [36]. Low TSH concentration may cause decreased degree of differentiation of thyroid epithelium, and lead to a predisposition for malignant transformation [51]. Under TSH stimulation, the mitotic activity of cells is increased, which then increase sensitivity to malignant tumors development [52]. TH and its receptor are related to cell growth and cancer progression [23]. At the integrin receptor for TH, T4 regulates cancer and endothelial cell division, tumor cell defense pathways (such as anti-apoptosis) and angiogenesis [24].

Our results showed that urinary MeP and EtP were associated with increased risk of benign nodules, and all three parabens were each associated with an increased risk of thyroid cancer in our single pollutant models. However, humans are simultaneously exposed to a complex mixture of pollutants from various sources [53]. Therefore, we applied a multi-pollutant model with three parabens and found that EtP

contributed most to the positive risk of thyroid cancer and benign nodules. Thus, our findings provided evidence to understand the effect of paraben mixture exposure through multiple routine on thyroid cancer and benign nodules.

In our study, urinary MeP and EtP were gender-specifically related to benign nodules, as well as EtP to thyroid cancer. We observed an increased risk of benign nodule in males but not female patients with urinary PrP levels above the median. Consistent with our results, a previous study has suggested that health damage like disrupt pregnancy caused by the estrogen-like effect of parabens may vary by gender [54]. Additionally, Gillies et al. found that biosynthesis and function of estrogens may differ by gender [55]. However, given that the number of female participants was approximately three times that of males in our study, further study is needed to verify the gender-specific mechanisms of parabens.

In our present study, the high detection frequencies for MeP, EtP and PrP were consistent with those presented in environmental monitoring study [56]. In addition, our results showed significant correlations between individual parabens, indicating similar sources and applications, which were in agreement with the results of existing studies [12, 14, 57]. The median concentrations of urinary MeP (29.30 μg/L) in our study were the highest, followed by PrP (0.99 μg/L) and EtP (0.54 μg/L), in line with a study conducted in Guangzhou and Shenzhen, South China [58]. However, the median concentrations (29.30 μg/L) of urinary MeP in our study were lower than that reported for in United States (63.5 μg/L), South Korea (39.7 μg/L) and Iran (69.06 μg/L) [59–61]; but higher than those reported for in Greece (11.6 μg/L), Belgium (16.1 μg/L) and Canada (25.45 μg/L) [12, 62]. Additionally, several studies have also presented the median concentrations of urinary EtP and PrP ranging from 1.0 to 12.4μg/L in adults, which were higher than those in our study [61–63]. The results of environmental and human monitoring study also showed that the Chinese population was less exposed to parabens than the American population [64]. The remarkable differences in the concentrations of urinary parabens between China and other countries may be attributed to the lower consumption rates of personal care products in China or differences in study population, geographical living environment and individuals [65]. Moreover, we found that women showed higher paraben exposure than men (P-value < 0.01). Several studies in the United States and Belgium also showed that urinary MeP and PrP concentrations in women were 3-4 times higher than those in men [12, 57, 62]. Such gender differences may be attributed to higher usage frequency of personal care products in women than that in men.

It is important to note that the samples included in the present study were collected before medical treatment and surgery, which provided an ideal opportunity to examine the relationship between paraben exposure, thyroid cancer and benign nodule. However, there are certain limitations which should be addressed in further studies. First, the sample size of our study was limited, which may lead to exposure misclassification. Second, urinary parabens show high variations in human population [66, 67]. One single point urine sample can produce exposure misclassification, suggesting repeated urine samples in further study to elevate exposure estimation. Third, because of the critical effect of iodine nutrition on thyroid, our results should be verified including urinary iodine as an important covariate in additional studies. Fourth,the most common histologic type of thyroid cancer is papillary thyroid cancer, we did not

analyze our data by type of thyroid cancer because of limit sample sizes. Thus, our results should be interpreted with caution. Finally, humans are often exposed to multiple environmental contaminants (e.g., heavy metals, phenols, pesticides, and flame retardant chemicals) simultaneously, further investigations looking into such multi-contamination is therefore encouraged to better elucidate the influence of co-exposure on thyroid disease.

5. Conclusion

In summary, we documented the paraben exposure levels before any medication or surgery, and estimated the relationships of environmental paraben exposure with the risk of thyroid cancer and benign nodules using a case-control study. We observed that urinary parabens increased the risk of benign nodules and thyroid cancer, when exposures evaluated as a mixture. In addition, we observed gender-specific correlation of individual parabens with the risk of benign nodules and/or thyroid cancer. Future longitudinal studies of the broader population are encouraging to further elucidate the underlying mechanisms.

Declarations

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Figures

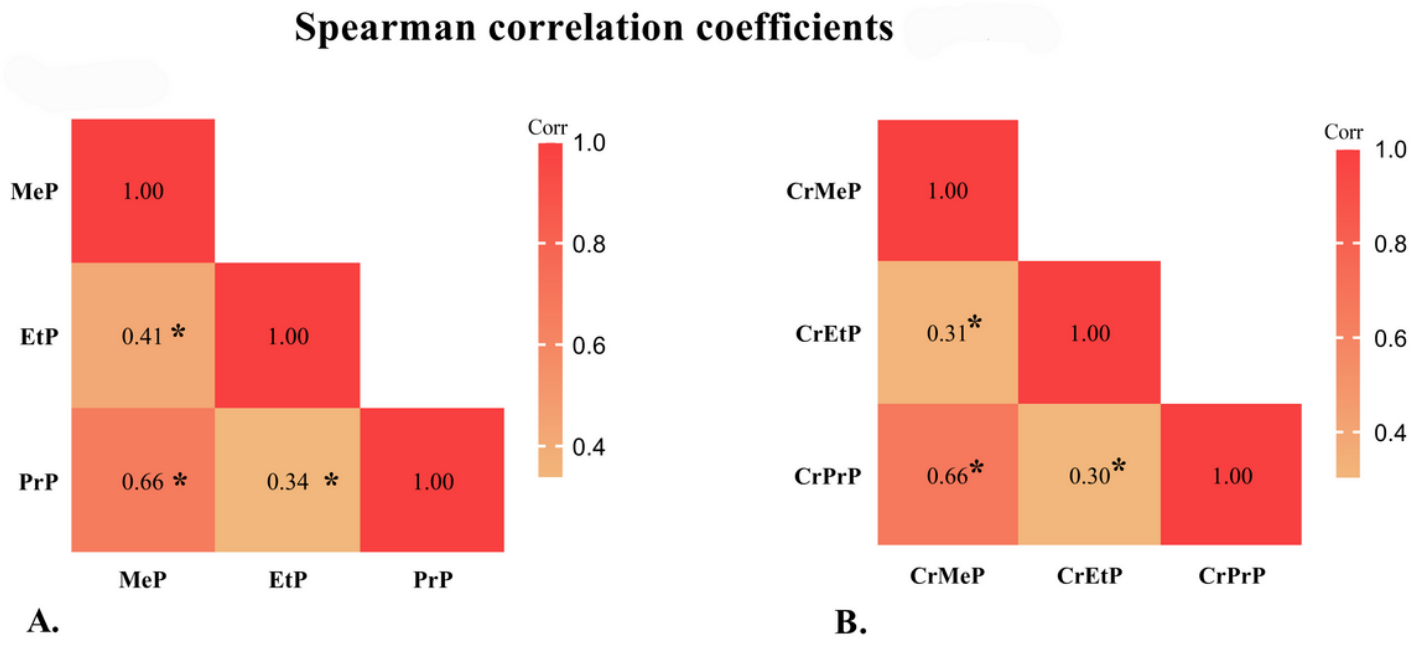

Figure 1

Spearman correlations between uncorrected urinary paraben concentrations (A) and creatinine-corrected urinary paraben concentrations (B). *, P-value < 0.05.

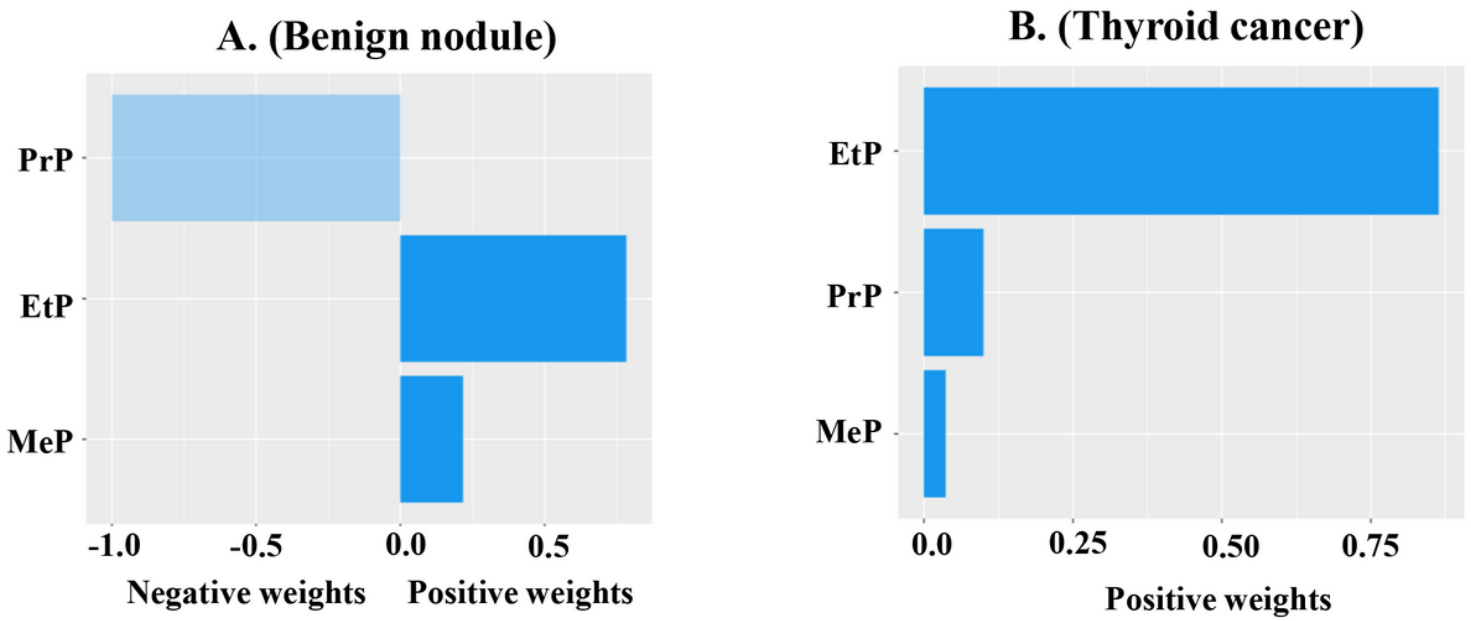

Figure 2

Weights representing the proportion of the positive or negative partial effect for each paraben biomarker on benign nodule (A) and thyroid cancer (B) in the quantile g-computation model. Creatinine-corrected urinary paraben concentrations were natural logarithm (ln)-transformed. The model was adjusted for gender, age, BMI, alcohol use, smoking status, and household income. The darker shading in the negative direction indicates overall mixtures effect in that direction. The length of the bars corresponds to the effect size only relative to other effects in the same direction.

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