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Relative Study on Blood BTEX, Testosterone Hormone, Kidney and Liver Functions in Gasoline Station Workers, Thailand

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Abstract

Benzene, toluene, ethylbenzene and xylene are volatile organic compounds (VOCs), usually referred to as BTEX. These compounds can affect human health upon its dose and time of exposures. VOCs are usually found in fuels and other solvents which commonly presented in the environment, however, little known about their effects on the endocrine system. This study aimed to evaluate the relationship between blood BTEX and testosterone hormone, and kidney and liver functions of gasoline station workers. The results showed average blood benzene, toluene, ethylbenzene and m-, p-xylene. o-xylene levels were 284.9, 201.3, 178.7, 35.9, 73.3 µg/L respectively. While average testosterone, blood urea nitrogen (BUN), creatinine, serum glutamic pyruvic transaminase (SGOT), serum glutamic pyruvic transaminase (SGPT) and alkaline phosphatase (ALP) levels were 13.4 nmole/L, 11.8 mg%, 1.0 mg%, 26.0 U/L, 30.9 U/L and 71.8 U/L respectively. Blood testosterone level was inversely related to toluene, m-, p-xylene, o-xylene and total BTEX levels (Linear regression analysis, $p<0.05$). In addition, testosterone level had strongly inverse-relationship to kidney function of BUN and creatinine (linear regression analysis, $p<0.05$ and $p<0.01$). In conclusion, this study supported that BTEX exposures were chronically affected the decreasing of testosterone level in reproductive system as well as to kidney function.

Keywords: BTEX, testosterone, liver, kidney, gasoline station.

Introduction

BTEX (benzene, toluene, ethylbenzene, and xylene), is a group of compounds which belonged to the broader category of volatile organic compounds (VOCs). Benzene is a known carcinogen[1], and has also been shown to cause blood disorders and impact the central nervous and reproductive systems[2]. Toluene may affect the reproductive system (including an increased incidence of spontaneous abortions) and central nervous system (CNS dysfunction depression and narcosis)[3-4].

Ethylbenzene and xylene may cause some respiratory effects such as throat irritation, chest constriction, irritation of the eyes, and neurological effects such as dizziness. Chronic (long-term) exposure to ethylbenzene by inhalation in humans has shown conflicting results regarding its effects on the blood, liver and kidney[5-8]. BTEX compounds can be emitted during various oil and gas operations activities, including flaring, venting, engines, produced water storage tanks, and during the dehydration of natural gas. The use of biomass energy has many unique qualities that provide environmental benefits as well as health affects[9]. The respective agencies in the government should build up the environmental awareness among people as other country[10].

Testosterone is one of six major androgen hormones made in the interstitial cells which stimulate secondary sex characteristics in males. it helps stimulate spermatogenesis in the testes (with FSH) and associates with sex drive[11-12]. Women's ovaries and adrenal glands share the responsibility for testosterone production for maintain muscle, ovarian function, bone strength, and contributes to sex drive or libido. Testosterone level of women is one-seventh the amount of testosterone each day that men do[13]. There were some evidences of solvent exposures on reproductive endocrine effects of lower preovulatory LH levels among women[14], lower prolactin and cortisol in cleaning task workers[15] and that may disrupt endocrine function in natural animal populations and humans[16]. Although endocrine disrupting chemicals (EDCs) have been know for many years, but this study has only recently attracted worldwide interest due to growing concerns about the relation of BTEX exposures and one of male sex hormones (testosterone) and its relations to kidney and liver function in gasoline workers.

Our object of this study was to study the relationship between blood BTEX and testosterone hormone, and between testosterone and kidney and liver functions of gasoline station workers who directly and continuously exposed to these VOCs.

Material and Methods

Population Study: A cross sectional survey was conducted by collecting 105 gasoline station workers (80 men and 25 women) of 11 gasoline stations located in Pathumwan area (figure-1), central Bangkok, Thailand. All subjects were healthy and had worked at the gasoline stations for more than six weeks. They

were provided with the consent form before the study. Permission to conduct the biological monitoring from human subjects in this study was approved by the Ethical Review Committee for Research Involving Human Research Subjects, Health Science Group, Chulalongkorn University.

Blood Sample Collection: Blood collection were conducted during their work shift (normally of 6-8 hours shift), using glass heparinize vacuum blood tube and stored at -20 ^{o}C before BTEX analysis.

Biological Analysis: The kidney function tests [blood urea nitrogen (BUN) and blood creatinine], liver function tests [alkaline phosphatase (ALP), serum glutamic oxaloacetic transaminase (SGOT), and serum glutamic pyruvic transaminase (SGPT)] and testosterone hormone were performed at Department of Clinical Microscopy, Faculty of Allied Health Sciences, Chulalongkorn University under standard laboratory quality control process.

Blood BTEX Analysis: The BTEX analysis was performed using the headspace-solid phase micro-extraction (HS-SPME) technique[17]. The quantity of blood benzene was analyzed under relative intensity of chromatographic signal for 40 minute. The Limit of Detections (LODs) of benzene, toluene, ethylbenzene and xylene were 10.00, 5.00, 20.00 and 10.00 µg/L (ppb) respectively and average coefficient of determination (r^2) was 0.9988286 for four chemicals (*m*-and *p*- xylene appeared with the same peak).

Statistical Analysis: All the statistical analyses were performed using the SPSS 17.0 for Windows Program. Descriptive statistic was used for the characteristics and blood biological levels of gasoline station workers. The relation between testosterone and BTEX levels and between testosterone and biological levels of kidney and liver function tests were analyzed by Multiple Linear Regression. All statistical significant value accepted at $p<0.05$.

Figure-1
Study area of Pathumwan area (• gasoline station)

Results and Discussion

Characteristics of Gasoline Workers: Of the total of 105 gasoline station workers, their mean of age, BMI and work period were 29.9 years, 23.2 kg/m^3 and 5.3 years, respectively (table 1). The general characteristics of age and work period of gasoline station workers were not difference between genders. There were 47.8% smoking and 98.0% drinking in men which were significantly higher than women (Chi-Square Test, *p<0.001*) while the BMI was significantly higher in women than in men (Independent t-test, *p<0.05*).

Blood Chemical Levels of Gasoline Workers: Most of blood chemical levels in men were significantly differences from women workers except blood benzene concentration (Independent t-test, table 2). Most of blood BTEX exposure levels were higher in women than in men workers, except benzene. It could be caused by the overall complexity of heart rate dynamics which are higher in women than men[18]. Blood benzene levels of gasoline station workers found in our study was 5.7 times of the BEI reference[19]. Testosterone, chemicals of kidney and liver function levels were also higher in men than women workers.

Relationship of Testosterone Levels and Characteristics of Gasoline Workers: The relations between testosterone level, BMI, smoking and drinking showed statistically significances at *p<0.01*, *p<0.05*, *p<0.05* respectively (table 3). Testosterone level was significantly inverse associated with BMI, which supported previous study of inverse correlation between testosterone and obesity[20]. There were high correlation of testosterone levels with body composition and its contribution to the balance of lipid metabolism. Moreover, testosterone was positively correlated to smoking and drinking of workers, as reported by Shiels et al[21].

Relationship of Testosterone Levels and BTEX Exposures: The relations between testosterone level, toluene, *m*-, *p*-xylene, *o*-xylene, total xylene and total BTEX statistically showed significances at *p<0.05*, *p<0.05*, *p<0.05*, *p<0.01* and *p<0.05* respectively (Linear regression analysis with adjusted for smoking, drinking, BMI and work period) (table 4). The testosterone levels were significantly inverse related to toluene, xylene and total BTEX. This result supported that toluene and xylene could affect testosterone level while benzene and ethyl benzene may be more specific to be genotoxicity at target organ including their toxicities of hematotoxicity and neurotoxicity[22-26]. However, high level of ethyl benzene can caused liver and kidney damage and has embryotoxic and teratogenic effects[6].

Table -1
Characteristics of gasoline workers

Parameter	Men N=80	Women N=25	Mean ± SD	*P-value**
Age (years)	29.4 ± 9.5	31.3 ± 8.7	29.9 ± 9.3	*NS*
BMI (kg/m^3)	22.5 ± 4.4	25.3 5.9	23.2 ± 4.9	*<0.05*
Work period (years)	5.1 ± 6.0	6.0 ± 7.6	5.3 ± 6.4	*NS*
Smoking N(%)	35 (43.8)	1 (4.0)	-	*<0.001*
Drinking N(%)	56 (70.0)	7 (28.0)	-	*<0.001*

* Significant difference between men and women workers

Table -2
Blood chemical levels of gasoline station workers

Blood chemical levels	Men N=80	Women N=25	Mean SD	*P-value**
Benzene (µg/L)	275.8 ± 88.2	314.1 ± 150.9	284.9 ± 106.9	*NS*
Toluene (µg/L)	175.0 ± 100.6	285.4 ± 164.9	201.3 ± 127.3	*<0.01*
Ethylbenzene (µg/L)	144.5 ± 99.1	288.1 ± 262.1	178.7 ± 164.6	*<0.05*
m-,p-Xylene (µg/L)	27.8 ± 26.1	61.9 ± 48.1	35.9 ± 35.6	*<0.01*
o-Xylene (µg/L)	55.7 ± 59.2	128.9 ± 126.6	73.3 ± 85.9	*<0.01*
Total Xylene (µg/L)	83.7 ± 84.3	174.5 ± 114.7	105.5 ± 99.8	*<0.01*
Total BTEX (µg/L)	684.7 ± 320.3	1062.1 ± 555.9	775.4 ± 419.5	*<0.01*
Testosterone (nmole/L)	16.3 ± 6.7	1.0 ± 0.9	13.4 ± 8.5	*<0.001*
BUN (mg%)	12.4 ± 3.1	9.8 ± 2.8	11.8 ± 3.2	*<0.001*
Creatinine (mg%)	1.0 ± 0.2	0.8 ± 0.1	1.0 ± 0.2	*<0.001*
SGOT (U/L)	28.1 ± 20.9	19.5 ± 5.8	26.0 ± 18.8	*<0.05*
SGPT (U/L)	34.0 ± 28.1	21.0 ± 10.7	30.9 ± 25.6	*<0.05*
ALP (U/L)	78.1 ± 40.7	51.6 ± 15.9	71.8 ± 38.0	*<0.01*

* Significant difference between men and women workers

Table -3
Relation between testosterone levels and the characters of gasoline station workers

Parameter	**Median**	**Reference**	**Linear regression analysis**			
			Standardized coefficients		**95% CI**	***P-value***
			B	Standard Error		
Testosterone (nmole/L)	M: 15.0 W: 0.8	M: 9.9-27.8 W: 0.2-2.9				
Age (years)	29.0	-	-0.119	0.110	-0.339 to 0.100	*0.283*
BMI (kg/m^3)	22.1	-	-0.177	0.055	-0.286 to -0.667	*0.002*
Work period (years)	3.0	-	-0.014	0.070	-0.153 to 0.125	*0.842*
Parameter	Median	Reference	Logistic regression analysis			
			Standardized coefficients		95% CI	*P-value*
			B	Standard Error		
Smoking N (%)	34.3	-	0.051	0.026	1.000 to 1.107	*0.052*
Drinking N (%)	60.0	-	0.058	0.026	1.000 to 1.116	*0.027*

Dependence variable: Testosterone

Table -4
Relation between blood BTEX and testosterone levels in gasoline station workers

Parameter	**Median**	**Reference**	**Linear regression analysis[a]**			
			Standardized coefficients		**95% CI**	***P-value***
			B	**Standard Error**		
Testosterone (nmole/L)	M: 15.0 W: 0.8	M: 9.9-27.8 W: 0.2-2.86				
Benzene (µg/L)	280.8	-	-0.010	0.008	-0.025 to 0.006	*0.218*
Toluene (µg/L)	185.8	-	-0.017	0.007	-0.030 to 0.003	*0.014*
Ethylbenzene (µg/L)	150.8	-	-0.010	0.006	-0.021 to 0.002	*0.096*
m-,p-Xylene (µg/L)	30.9	-	-0.059	0.025	-0.109 to -0.010	*0.019*
o-Xylene (µg/L)	43.3	-	-0.026	0.010	-0.047 to -0.006	*0.013*
Total Xylene (µg/L)	75.2	-	-0.025	0.009	-0.042 to -0.008	*0.004*
Total BTEX (µg/L)	713.5	-	-0.005	0.002	-0.009 to 0.000	*0.019*

Dependence variable: Testosterone [a]Adjusted for smoking, drinking, BMI and work period

Table -5
Relation between testosterone and kidney and liver functions in gasoline station workers

Parameter	**Median**	**Reference**	**Linear regression analysis[a]**			
			Standardized coefficients		**95% CI**	***P-value***
			B	**Standard Error**		
Testosterone (nmole/L)	M: 15.0 W: 0.8	M: 9.9-27.8 W: 0.2-2.9				
BUN (mg%)	12.0	6.0-20.0	0.646	0.263	0.124 to 1.168	*0.016*
Creatinine (mg%)	0.9	0.5-1.5	12.436	4.128	4.237 to 20.634	*0.003*
SGOT (U/L)	21.0	<40	3.455E-02	0.045	-0.055 to 0.124	*0.446*
SGPT (U/L)	24.0	<40	4.126E-02	0.035	-0.028 to 0.111	*0.240*
ALP (U/L)	64.0	26.0-117.0	-1.10E-02	0.024	-0.058 to 0.036	*0.643*

Dependence variable: Testosterone [a]Adjusted for smoking, drinking, BMI and work period

Relationship of Testosterone Levels and Liver and Kidney Functions: The testosterone levels were positively and strongly related to kidney function of BUN and creatinine at $p<0.05$ and $p<0.01$ but not related to liver function (table 5). In addition, testosterone level was also found to be significantly related to kidney function of BUN and creatinine as well as blood BTEX levels by using linear regression analysis. Similar result was done in male rat by Muraoka[27]. He found the correlation between the serum concentration of total and free testosterone and each parameter of renal function by using linear regression analysis and testosterone replacement affected the deterioration of renal function in middle-aged rats.

However, several reports suggested that testosterone has a stimulatory effect[28-29] such as testosterone increases collagen synthesis by vascular smooth muscle cells in culture[28], administration of testosterone increases the accumulation of collagen and elastin in the aorta of normal and cholesterol-fed animals[31].

Conclusion

This study revealed that BTEX exposures were chronically affected the decreasing of testosterone level in reproductive system which depended on dose and duration of exposure. But at present study, the testosterone level was still in normal range. It should be followed up of these workers. Female workers may be more affected than male workers cause of higher blood BTEX levels. In addition, testosterone level had relation to kidney function.

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Abbreviations

BTEX: Benzene, Toluene, Ethylbenzene and Xylene
BUN: Blood urea nitrogen
Cr: Creatinine
SGOT: Serum Glutamic Oxaloacetic Transaminase
SGPT: Serum Glutamic Pyruvic Transaminase
ALP: Alkaline Phosphatase
CI**:** Confidence Interval
ACGIH: American Conference of Governmental Industrial Hygienists
ATSDR: Agency for Toxic Substances and Disease Registry
NIOSH: National Institute for Occupational Safety and Health
US EPA: US Environmental Protection Agency

Conflict of Interests: This study was none conflict of interest

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