# Environmental Exposure to Volatile Organic Compounds in Indoor Chemical Laboratories: A Comparative Study of BTEX Characteristics through Monitoring Strategies

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**ABSTRACT**

BTEX (benzene, toluene, ethyl benzene and xylenes) are the most important categories of VOCs that occur in the indoor air and often used in chemical laboratories. In the present study the excretion of urinary BTEX were evaluated as biomarkers of exposure to these compounds. The mean value of benzene in breathing zone and the total benzene uptake during the work shift of the two groups of technicians and students in chemical laboratories were 32.11and 46.82 $\mu g\ m^{-3}$ and 14.55 and 34.11ng$L^{-1}$, respectively, which were significantly greater than the occupationally non exposed groups. Good correlations ($0.839 \leq r \leq 0.946$) between the mean values of BTEX in breathing zone and the urinary concentrations were observed.

**Keywords**: BTEX; Exposure; Urinary levels.

## INTRODUCTION

Air pollution is one of the major health impacts in modern communities and people can be exposed by several ways (e.g. ambient air, indoor air) which are responsible for many diseases. Indoor air quality represents a major concern since more than three decades. Most of people spending approximately 90% of their times indoors. Some studies showed that indoor concentrations of many volatile organic compounds (VOCs) were greater than those outdoors[1]. BTEX (benzene, toluene, ethyl benzene and xylenes) are the most important categories of VOCs that occur in the indoor air. People exposed to BTEX, mainly in workplaces (industries, laboratories, hospitals, and others) have an increased probability of acquiring degenerative diseases. These substances target the central nervous system and are easily absorbed by the human organism through the lungs and, in some cases, through the skin[2-4]. BTEX are used widely in organic synthesis and analysis procedure as reagents and solvents. In the chemical laboratories, vapor of these kinds of chemicals are a common cause for indoor pollution. The technician in chemical laboratories, which can be considered to be 'special indoor places', as well as students who worked on their thesis are exposed to high concentrations of BTEX. The main route for human exposure to BTEX is via inhalation, but the compounds are also adsorbed by mouth and skin contact[5]. For the assessment of occupational exposure to BTEX determination of compounds per se in breathing zone and in urine was proposed[6]. According to literature, in estimation of workers' exposure with biological samples, analysis of

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the concentration of unchanged metabolites in urine detected after the work shift seems to give the most reliable estimation of exposure[7,8]. The aim of this study was to determine the exposure levels for technician and students in chemical laboratories during routine work shift, by environmental and biological monitoring to provide information on the potential health impacts.

## MATERIALS AND METHODS

*Study Population*

One hundred twenty healthy men from the city of Tehran, Iran, were enrolled in this study. The study population included 40 chemistry laboratory technicians from several universities and 40 students who work in the same laboratories in Tehran as an occupationally exposed groups and for the measurement of any background levels originating from other sources such as ambient air, urine samples were collected from 40 occupationally nonexposed persons in the same organization acted as referents. All of the subjects were men between 20 and 43 (mean=28) years old and none of them were smokers. In order to dermal protection, it was requested from laboratory technicians and students to wear plastic gloves during work shift. *Analytical determinations*

*Environmental monitoring*

Radiello passive samplers were used for measuring personal exposure to BTEX[9,10]. In our study, Radiello® samplers were attached to a distance of ~10 cm from the operator's face, to the right or to the left depending on whether the operator was right handed or left handed. After exposure, the chemical substances collected on sorbing cartridges were desorbed into the storing glass tube for about 30 min with 2 mL of benzene free carbon disulfide in the presence of the internal standard solution containing benzene-d6. Determinations were performed by means of gas chromatography (Agilent GC/MS 6890/5973detector; HP-5, 30 m × 0.25 mm × 1 μm). The instrumental temperatures were as follows: injector temperature, 250ºC; initial oven temperature, 40 ºC (held for 10 min), increased to 90 ºC at a rate of 10 ºC $min^{-1}$, held for 3 min, and

then to 120°C at 20 °C min−1, hold 2 min, and then to 160 °C at 30 °C min−1, (final temperature 2 min hold). The inlet was operated in splitless mode. The acquisition mode was SIM. Quantification limit (LOQ) was 1 µgL$^{-1}$ for all the analytes. Air concentration (µg m$^{-3}$) was calculated using the equation:

$$\frac{m}{Q(L/\min)\cdot t(\min)}$$

where m = mass of analytes determined in desorbing solvent; Q = uptake rate of substances (80 mL min$^{-1}$ for benzene; 74 mL min$^{-1}$ for toluene; 68 mL min$^{-1}$ for ethylbenzene; 70 mL min$^{-1}$ for m-xylene; 65 mL min$^{-1}$ for o-xylene and 70 mL min$^{-1}$ for p-xylene); t = exposure time[11]. *Biological monitoring*

*Sampling*

Urine samples were collected at the beginning and at the end of the shift. In order to avoid loss of analytes during collection and storage, urine samples (0.6 ml) were immediately transferred in 2 ml SPME glass vials containing 300 mg of NaCl. The samples were shaken and stored at −20 ◦C until analysis. *Analysis of urine samples*

BTEX were analyzed using head-space gas chromatography and equipped with single quadrupole mass detection (GC-MS). The sampling procedure involved placing 0.6 ml of sample into a 2 mL vial containing 300 mg NaCl and sealing with a screw-top septum containing cap. The sample was then spiked with a 50µL of methanol containing 1µgmL$^{-1}$ of benzene-d as internal standard. The vial was stirred in a Vortex mixer for 3 min, and then placed in a water bath maintained at 35±0.1 ◦C for 5 min to establish phase equilibrium. The vial and SPME holder were clamped into a stand that allowed the vial to be immersed in the water bath only up to the level of the liquid in the vial. The SPME needle was then inserted through the septum into the headspace so as to locate the tip of the exposed PDMS 100 µm fiber approximately 0.5cm from the top of the liquid; the fiber was allowed to equilibrate for 8 min. The fiber was then retracted, removed from the vial, and placed immediately into the injector of the GC system. The SPME holder was adjusted so that the exposed fiber tip was positioned about halfway (1.5 in.; 1 in. =2.54 cm) into the GC injector port liner when extended from the protective needle. Injection was accomplished by extending the fiber in the heated inlet for 3 min, and the splitter was opened after 1min. The additional 1.5min of exposure time in the injector port allowed the fiber to be cleaned of any compounds that were not desorbed in the initial 3min. Blank samples containing internal standards were analyzed at the beginning and at the end of the sample queue. Triplicates of each sample were extracted by the SPME technique (11). Determinations were performed by means of gas chromatography (Agilent GC/MS 6890/5973detector; HP-5, 30 m × 0.25 mm × 1 µm). The instrumental temperatures were as follows: injector temperature, 250ºC; initial oven temperature, 40 ºC (held for 3 min), increased to 70 ºC at a rate of 3 ºC min$^{-1}$, held for 1 min, then increased to the final temperature 250 ºC at a rate of 20 ºC min$^{-1}$, where it was held for 5 min. The inlet was operated in splitless mode. The temperature of the transfer line was maintained at 290 ºC. Helium (99.99%) was used as carrier gas at 1ml min$^{-1}$ (constant flow). The source and quadrupole temperatures were kept at 230 and 150 ºC, respectively. The electronic beam energy of the mass spectrometer was set at 70 eV. The mass selective detector was operated in electron impact (EI) mode using selected ion monitoring (SIM). The LOQ values for all compounds were 15 ngL$^{-1}$.

*Statistics*

Mean urinary concentrations of BTEX among three groups (laboratory technicians, students and occupationally nonexposed persons) were analyzed and because the distribution of data was not normal; the analysis was carried out by means of two statistical procedures: analysis of variance (one way ANOVA) followed by scheff's post hoc test and Kruskal-Wallis test. Results were expressed as mean ± S.E and 95% confidence intervals. The level of significance was set to 0.05 and P values >0.05 were assumed to be nonsignificant.

## RESULTS AND DISCUSSION

The toxic effects of BTEX on the human nervous system have been known for some time[12]. Benzene was shown to be carcinogenic to multiple organ sites in both sexes of multiple strains and multiple species of laboratory animals exposed via various routes[13]. The most important source of human exposure to BTEX is from breathing of contaminated air and through cigarette smoke[14]. Exposure to BTEX from water contributes only a small percentage of the total daily intake, compared with inhaled air[15]. In this study, we used the diffusive personal samplers for monitoring of BTEX in breathing zone because they were small, silent, and easy to handle and did not need any field calibration. Since there may be a significant risk of dermal exposure, it was requested from persons to wear plastic gloves during the work shift, so no significant skin exposure occurred during the study. Table 2 shows the results of environmental measurements in the three selected groups during a day work shift. The mean value for exposure of BTEX in breathing zone of occupationally exposed groups were significantly greater than the occupationally non exposed group but below the 2000 ACGIH TLV (TWA)[16]. Toluene was found to be the most abundant component, followed by xylenes (o,m and p) in breathing zone of all groups. Since toluene, is the common solvent which is used in chemical lab in

Table 1: The mean value (SD) for exposure of BTEX in breathing zone.

| Compounds<br>Groups | Benzene | Toluene | Ethyl benzene | o-xylene | m-xylene | p-xylene |
|---|---|---|---|---|---|---|
| laboratory technicians | 32.11±12.64 | 554.65±147.99 | 11.48±4.65 | 47.48±14.48 | 33.78±11.98 | 48.53±21.85 |
| Students | 46.82±15.57 | 685.22±169.61 | 15.29±1.67 | 53.90±14.51 | 52.03±18.45 | 53.84±24.47 |
| occupationally nonexposed persons | 12.01±6.21 | 82.12±57.18 | 7.45±0.61 | 23.29±8.42 | 29.89±8.32 | 19.16±12.91 |
| P value* | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 |
| P value** | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 | <0.001 |

* one-way ANOVA
** Kruskal-Wallis test

Iran, airborne toluene usually occurs at higher concentrations than other compounds.

The concentration of benzene detected in in breathing zone of occupationally exposed groups was significantly lower than permitted standard concentration, maybe because of general awareness about benzene serious side effects on health and being more cautious in working with benzene. Airborne BTEX levels of chemical lab were similar to the ones related to other jobs which have occupational exposure to BTEX asserted in the literature[17,18].

The results of the mean urinary concentrations of BTEX in all groups in two different samples collected: before starting work and at the end of the shift are shown in Table 1.

In order to search the relationship between BTEX in the air of workplace and in urine, regression analyses were performed. The results are shown in Fig 1. Time weighted average of environmental concentration (in the breathing zone) ($C_{env}$) and the urinary concentration ($C_{urine}$) of BTEX were regressed well. Previous studies indicated that BTEX metabolites in urine are preferred biological index for monitoring BTEX exposures[16,19]. In general the advantage of using unmetabolized compounds as biomarkers is that the urinary concentration of the unmetabolized substance is less influenced by inter individual metabolic differences than the urinary disposition of corresponding metabolites. However, due to the similar significance of unmetabolized chemicals in blood and urine as biomarkers of exposure[20], and also since there is good correlation between $C_{env}$ and $C_{urine}$, for the assessment of exposure to BTEX, the urinary concentrations of BTEX after the 1 day shift were chosen as an indicator of a day's (acute) exposure. The mean BTEX concentrations before starting work and at the end of the shift in laboratory technicians and students were significantly greater than the occupationally non exposed group. The total benzene uptake during the work shift in two groups of laboratory technicians and students were calculated to be on average 16.95 and 28.30 ng $L^{-1}$, respectively. The concentration of benzene increased during the day for both groups of occupationally non exposed. The increase is more marked among students who work directly with different solvents. As can be seen in Table 2, excretion of BTEX in occupationally non exposed group was significantly lower than occupationally exposed workers ($P<0.001$) and their urinary concentration was approximately constant during

Table 2: Mean urinary concentration (SD) of BTEX in three groups before starting work shift and at the end of work shift.

| Compounds | Benzene | | Toluene | | Ethyl benzene | | o-xylene | | m-xylene | | p-xylene | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Sampling time | Before | After | Before | After | Before | After | Before | After | Before | After | Before | After |
| laboratory technicians | 35.16±11.28 | 49.71±21.36 | 91.23±15.51 | 122.31±26.15 | 21.71±10.31 | 25.76±9.68 | 41.32±28.12 | 52.73±21.64 | 57.61±26.13 | 70.12±32.15 | 61.71±24.14 | 73.45±15.26 |
| Students | 44.56±21.32 | 78.67±15.62 | 101.71±36.12 | 174.13±35.26 | 29.31±10.25 | 36.71±14.69 | 47.71±28.47 | 58.12±36.25 | 62.31±40.15 | 78.25±28.39 | 65.13±34.58 | 81.29±28.94 |
| occupationally non-exposed persons | 20.61±9.25 | 21.54±15.26 | 54.15±36.85 | 56.71±28.94 | 17.21±12.36 | 18.01±11.42 | 25.12±18.58 | 26.37±19.28 | 31.24±15.74 | 32.11±19.15 | 29.12±15.86 | 30.22±20.18 |
| P value* | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 |
| P value** | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 | <0.05 |

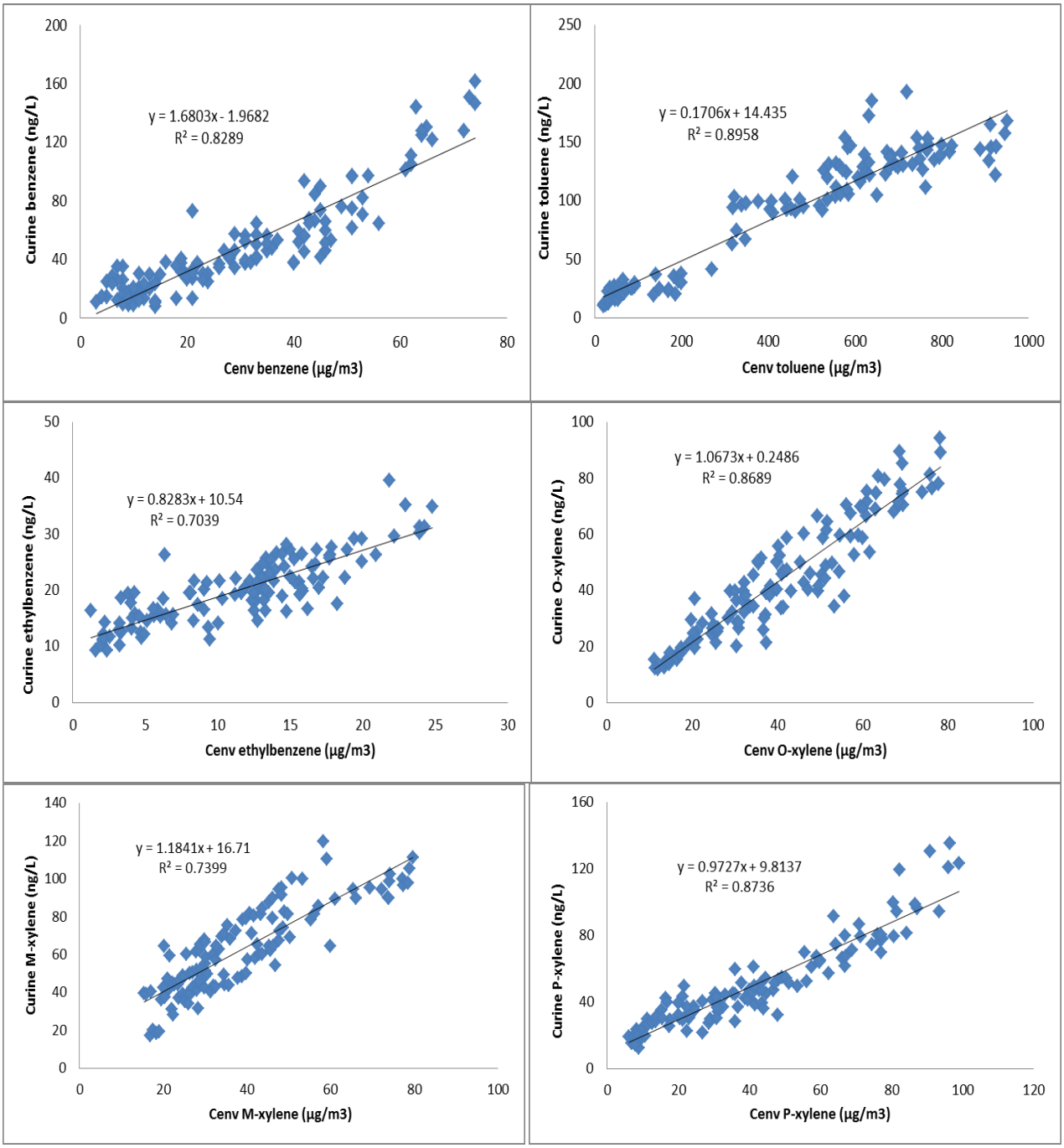


Figure 1: Scatter diagram relating the time—weighted average of environmental concentration (in the breathing zone) ($C_{env}$) and the urinary concentration ($C_{urine}$) of BTEX.

one work shift. BTEX have been widely used in many chemical laboratories in the entire world, in organic synthesis and analysis procedure as reagents and solvents. As the results indicated, a significant difference was observed in BTEX, concentrations in both pre and postshift samples among job categories ($p<0.05$ by ANOVA and Kruskal–Wallis test). The mean BTEX, concentrations in exposed workers were significantly greater than that of occupationally non exposed group. The urinary BTEX levels detected in this study was less than those were obtained for workers who work in paint and footwear factory[20]; whereas urinary BTEX levels is much more than levels which are reported for American bus drivers[16].

Because of serious health adverse effects of organic solvent, monitoring of these compounds in occupational environments is necessary, so it seems that in estimation of workers' exposure with biological samples, according to this study, analysis of the concentration of BTEX detected before and/after the work shift seems to give the most reliable estimation of acute exposure. In conclusion, from the results of this pilot study it seems that indoor air in chemical labs of universities is in an acceptable condition

but it should be remembered that little is known about the effects to human health of chronic exposure to 'low' doses of BTEX.

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