Ecotoxicology and Environmental Safety 197 (2020) 110615

Contents lists available at ScienceDirect

Ecotoxicology and Environmental Safety

journal homepage: www.elsevier.com/locate/ecoenv

The exposure risk of typical VOCs to the human beings via inhalation based on the respiratory deposition rates by proton transfer reaction-time of flight-mass spectrometer

Yi Yang, Hao Luo, Ranran Liu, Guiying Li*, Yingxin Yu**, Taicheng An

Guangdong Key Laboratory of Environmental Catalysis and Health Risk Control, Guangzhou Key Laboratory Environmental Catalysis and Pollution Control, School of Environmental Science and Engineering, Institute of Environmental Health and Pollution Control, Guangdong University of Technology, Guangzhou, 510006, China

ARTICLE INFO

Keywords:
VOCs
Inhalation exposure
Respiratory deposition rates
PTR-TOF-MS
Conventional lung function

ABSTRACT

The respiratory deposition rates are the important analytical parameters for human health risk assessment related to the environmental volatile organic compounds (VOCs). In present study, the deposition rates from the linear regressions of CH_2O, CH_5N, C_2H_6O, $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8, C_8H_8, and C_8H_{10} of 120 healthy volunteers were obtained with significantly different from the respective calculated deposition rates. The CH_2O (formaldehyde) has the highest deposition rate, indicating the highest associated exposure risk of CH_2O if the persons are exposed to the same concentrations of these VOCs through inhalation. In order to explore the effects of the breathing models and sampling time on the deposition rates of VOCs, volunteers were first asked to breathe successively with nasal-in-nasal-out, oral-in-nasal-out, and oral-in-oral-out breathing models before and after three meals for three days. Sampling time variation has no effect on the deposition rates of selected VOCs, while the deposition rates of $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8 and C_8H_{10} by nasal-in-nasal-out were significantly different from oral-in-oral-out and nasal-in-oral-out models. Among all the breathing models, nasal-in-oral-out comprises the entire respiratory system. In order to further validate the results, the deposition rates of the selected VOCs were calculated in 120 healthy volunteers using nasal-in-oral-out breathing model for unlimited time after the conventional lung function examination. Difference in gender and body mass index had no effect on the deposition rates of VOCs, while the age affects the deposition rates of CH_2O, CH_5N and $C_2H_4O_2$. Positive correlation analysis between lung function factors and deposition rates revealed that the individuals with larger lung function factors are more susceptible to deposit the VOCs. Overall, the main conclusion can be drawn that the respiratory deposition rates were influenced by the physiological factors. Therefore, the major objective for future research is to accurately calculate the deposition rates of environmental VOCs for health-risk assessment.

1. Introduction

Volatile organic compounds (VOCs) are commonly present in the environment (Liang et al., 2020; Yang et al., 2019). Ingestion, inhalation and dermal exposure to VOCs can induce many adverse health effects (Cao et al., 2018; Weisel and Jo, 1996). Inhalation is the major route of exposure because of the volatile property of VOCs. Previous studies have demonstrated that the respiratory exposure of VOCs via the inhalation route is associated with the risk of specific diseases, including asthma (Rumchev et al., 2004; Tagiyeva and Sheikh, 2014), chronic obstructive pulmonary disease (COPD) (Audi et al., 2017), cardiovascular diseases and various cancers (Lewtas, 2007; Li et al., 2019; Omidi et al., 2019; Xu et al., 2009). However, not all the inhaled VOCs are deposited in the respiratory system. Therefore, the percentage of the inhaled toxic VOCs deposited and absorbed inside the body play an important role in health risks (He et al., 2019). The deposition of the VOCs in the lung is related to the respiratory deposition rates of VOCs or bioavailability (Wei et al., 2018). The default value of 100% is used to assess the risk of VOCs, while the retention of VOCs in lungs varies from less than 20% to more than 90% (Jakubowski and Czerczak, 2009).

Therefore, it is essential to measure the accurate deposition rates of the VOCs in the respiratory system, since most of the deposition rates of VOCs are usually calculated for the risk assessment.

According to the deposition process, it is possible that the deposition rate of a pollutant is affected by both individuals and substances

* Corresponding author.
** Corresponding author.
E-mail addresses: ligy1999@gdut.edu.cn (G. Li), yuyingxin@gdut.edu.cn (Y. Yu).

https://doi.org/10.1016/j.ecoenv.2020.110615
Received 1 February 2020; Received in revised form 6 April 2020; Accepted 8 April 2020
Available online 20 April 2020

(Beauchamp, 2011). Different research studies have reported that the VOCs' deposition rates are also affected by the concentrations of VOCs in the inhaled ambient air. Huang et al. found that the respiratory deposition rates of benzene, toluene, ethyl benzene and xylene decreased in a logarithmic way when exposure levels of aromatic hydrocarbons (AHs) decreased (Huang et al., 2017). Spaněl et al. reported the linear relationship of seven selected VOCs with exhaled and inhaled VOC concentrations in the air (Spaněl et al., 2013). Unfortunately, the changes in deposition rates of VOCs determined by subjects is insufficiently explored. So far, Huang et al. found that physiological parameters including gender and body mass index or body fat ratio are not the dominant factors influencing the deposition rates of AHs (Huang et al., 2017). Many studies have been conducted to emphasize the effect of subjects on the deposition rates of VOCs. For example, the change in the breathing models immediately affects the exhaled VOC concentrations (Sukul et al., 2017a). Although the change in the exhaled ethanol level was not statistically significant, the levels after half an hour of both breakfast and lunch had higher tendency as compared with the overnight fasting concentrations (Bikov et al., 2013). Similarly, Martinez-Lozano Sinues et al. found that the 36–49% of 111 m/z channels in the breath of all individuals showed significant circadian variation in at least one individual (Martinez-Lozano Sinues et al., 2014). Sukul et al. observed the forced expiratory volume (FEV) maneuver induced changes in the concentrations of VOCs and alveolar isoprene increased by 6% and 21% respectively during maximum exhalation process (Sukul et al., 2016). Overall, previous studies have almost exclusively focused on the influencing factors of the human exhaled VOCs, However, the research in the deposition rates and the retention of VOCs via inhalation remains limited.

Respiratory deposition rate is an important parameter to evaluate the harmful effects of VOCs on the human health. The change in the exhaled gas concentration is regarded as the phenotype, and changing the deposition rates is one way of describing the risk to human health. Furthermore, the effects of individual human physiology on deposition rates predominantly depends on the gas exchange and human metabolic rate. The physiological factors such as gender, age, and the body mass index (BMI) regarding to human metabolic condition potentially affect the respiratory deposition factors (Huang et al., 2016, 2017). Pulmonary ventilation and capacity represented by lung function factors influence the alveolar gas exchange and thereby, the sink of the VOCs (Sukul et al., 2016, 2017b). Therefore, the respiratory exposure methods, circadian variation and individual human physiological conditions should be explored to the effect of the deposition rates of VOCs.

In this study, two healthy female and two male volunteers were firstly asked to breathe in three different ways including nasal-in-nasal-out, oral-in-nasal-out, and oral-in-oral-out successively in a normal laboratory. They were tested before and after each meal for three days using a homemade real-time breath sampling device coupled with a proton transfer reaction-time mass spectrometry (PTR-TOF-MS). Real-time respiratory deposition rates were measured as the volunteers were exposed to indoor VOCs at several parts per billion (ppb) under the normal allowable concentration. Furthermore, 120 volunteers were asked to be observed in the same laboratory as the former experiment using buffered exhaled tube coupled with the PTR-TOF-MS. The purposes of this study are to (1) find the relationship between the deposition dose and the exposure levels of different VOCs, (2) check if breathing models and circadian variation can influence the respiratory deposition rates of VOCs, and (3) elucidate the physiological factors, including gender, age, BMI and conventional lung function parameters affecting the deposition rates.

2. Materials and methods

2.1. Subjects

Before the experiment, all the volunteers signed the informed consent, and all the investigations were carried out in a normal laboratory. The gas inhaled by the subjects is the normal ambient air in the laboratory determined according to the described method (Huang et al., 2017). In order to check whether the breathing models and sampling time could possibly influence the deposition rates of VOCs, four healthy volunteers (equal female to male ratio) were firstly invited to collect the inhaled and exhaled gas using a self-exhaled gas collection device, and then measured with PTR-TOF-MS (Ionicon Analytik GmbH, Innsbruck, Austria). Volunteers were asked to breathe through three different models, including nasal-in-nasal-out, oral-in-nasal-out, and oral-in-oral-out successively in each test. In addition, sampling time was determined six times before and after each meal in the morning, noon and evening for three days.

Furthermore, a total of 120 healthy volunteers (Female: Male = 5: 7) were recruited to find the relationship between human physiological parameters and the deposition rates of the selected VOCs. All volunteers were asked to complete a questionnaire regarding gender, age, height, weight, BMI, smoking, drinking alcohol consumption and present health. All the obtained information is provided in Table S1. In addition, pulmonary function test of the volunteers was performed using the Masterscreen Pneumo (Micro lab, Jaeger, Germany), and the concentrations of the breathing VOCs were measured using the Buffered End-Tidal (BET) in combination with PTR-TOF-MS in the same laboratory for the following experiment.

2.2. The breathing sampling device

In the first experiment, a homemade breathing device was used to determine the difference in the breathing models and the circadian variation impact on the respiratory deposition rates of VOCs. However, for the rapid measurement of the large quantity of the exhaled gas, commercially available BET was used in the following experiments. A schematic of the real-time exhaled gas collection device is depicted in Fig. S1. This device mainly consists of a buffer gas bag connected with three stainless steel tubes. The main body of the device is a Teflon buffer gas bag placed in a heating box incubator set at 37 °C. There are three-way check valves connected to the environment air inlet tube, buffer gas bag and breathing tube. The breathing tube attaches the face mask to facilitate the nasal-in-nasal-out or oral-in-nasal-out breathing in the subjects. However, the connection of breathing tubes with a mouthpiece enabled the oral-in-oral-out breathing. The check valve in the environment air inlet tube was set to prevent the ambient gas entering the bag. In addition, another valve is provided at the air inlet of the gas bag in case the gas in the gas bag is sucked up. Another three-way valve connects the sample tube, the gas pump and the gas bag. The exhaled gas was drawn into the PTR-TOF-MS to quantitatively analyze the concentrations of VOCs. The residual gas in the bag was drained out of the buffer gas bag using pump via the three-way valve. During the sampling process, the exhaled gas of the human body was buffered in the buffer gas bag. Then, the resultant buffered gas was drawn into the PTR-TOF-MS with the sample tube, and the excess gas was squeezed out by the next exhaled gas. After each examination, the buffer gas bag was cleaned by filling it with high-purity nitrogen (99.999%). The residual gas was evacuated with a gas pump, and the cleaning steps were repeated for three times. A heating box conserves the 500-mL buffer gas bag at 37 °C to avoid the condensation of humid exhaled air and the adsorption of the target compounds.

2.3. Lung function measurements

In the second experiment, each subject was required to pass the routine lung function measurement using the Masterscreen Pneumo (Micro lab, Jaeger, Germany). The lung function factors, including Forced Expiratory Volume in 1 s (FEV1), Forced Vital Capacity (FVC), Maximum Mid Expiratory Flow rate (MMEF), and Maximal Ventilator Volume (MVV), were measured according to the recommended

guidelines (Miller et al., 2005).

A commercial PTR-TOF-MS 1000 was utilized to measure the concentrations of the selected VOCs present in the atmospheric air and the breathing samples. The samples were directly extracted into the reaction drift tube of the PTR-TOF-MS through the heated (at 80%) polyetheretherketone tubing. The drift tube was operated at the voltage of 600 V, pressure of approximately 2.30 mbar, and the temperature of 80 °C with an E/N ratio of approximately 134 Townsend (Td) (where E is the electric field strength and N is the number density of a neutral gas; 1 Td = 10–17 V cm^2). The PTR-TOF-MS was operated using protonated water (H_3O^+) as the reagent ion within the mass range of m/z 18–240.

Before sampling, the PTR-TOF-MS was calibrated by the standard sample TO-15 (Linde Spectra Environment Gases, USA). The C_6H_6, C_7H_8, C_8H_8, C_6H_5Cl, $C_6H_4Cl_2$ and $C_6H_3Cl_3$ were selected to calculate the transmission values. The transmission values improved the mass-dependence of the actual mass separation and the detector in the transfer system according to the manufacturer's instructions. The transmission function was entered into TOF-DAQ (Tofwerk AG, Switzerland). The ppb concentrations of each VOC were calculated using the correlation described previously from the signal intensities (cps) of raw data in TOF-DAQ (Tofwerk AG, Switzerland) (Li et al., 2013; Lindinger et al., 1998). The instrument preparation methodology is mentioned in a previous study (Han et al., 2019). The R^2 of the calibration curve was 0.9951 (Fig. S2). The detection limits were determined by a previously descripted method (Huang et al., 2016), with values ranged from 0.015 to 0.021 ppbv for typical nine kinds of VOCs, including CH_2O, CH_5N, C_2H_6O, $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8, C_8H_8 and C_8H_{10}. In addition, the background values of the instrument were measured by introducing high-purity nitrogen (99.999%) before each experiment. The internal calibration of PTR-TOF-MS spectra adjusted the dead time, based on m/z = 21.022 ($H_3{}^{18}O^+$), m/z = 37.027 (cluster H_3O^+) and m/z = 59.049 ($C_3H_7O^+$) (Yuan et al., 2017).

Ambient air, which was the air inhaled by subjects, was also experimented using the breathing sampling device in the first experiment and the buffered exhaled tube in the second experiment. It was then measured with PTR-TOF-MS. According to the operating steps as described above, the inhaled and exhaled air for each subject was continuously measured at least for 10 s to determine the respiratory deposition rates of the selected VOCs contained in these gases. All the experiments were conducted in the laboratory with numerous instruments without any chemical storage. Before the breathing test, each subject breathed at least seven times in the laboratory to make sure the exhaled gases were produced by inhaling the ambient air. In each examination, the ambient air was measured until the VOCs' response signals remained stable for at least 200 cycles. The concentrations of VOCs in ambient air at the stable stage were regarded as the levels of the inhaled air. During the subsequent measurement of exhaled gas, each subject was asked to breathe at normal frequency and depth to ensure the VOC response signals remained stable for at least 200 cycles. In the first experiment, the subjects were requested to breathe by three ways of breathing, including nasal-in-nasal-out, oral-in-nasal-out and oral-in-oral-out successively. According to the results of the first experiment, the method of oral-in-nasal-out was preferred to applying in the second experiment. The deposition rate is defined as Eq. (1).

$$K = \frac{C_i - C_e}{C_i} \quad (1)$$

where, C_i (ppb) is the concentration of the target compound in the inhaled air by the test subject, and C_e (ppb) is the concentration of the target compound in the exhaled air. ($C_i - C_e$) is the mean of the amount of the target compound absorbed by the subject (Huang et al., 2016).

2.4. Statistical analysis

The Student's *t*-test in the statistical software package SPSS (version 19) was used to examine the statistical differences in the deposition rates of VOCs according to the different breathing models and sampling time classifications. In addition, the Mann-Whitney *U* test was used to find the statistical differences in the deposition rates of VOCs in different individuals of gender, age and BMI. Two-tailed tests of significance were also used, where $p < 0.05$ indicates statistical significance. It was also used as a critical value for significant correlation.

3. Results and discussion

3.1. Deposition rates of the selected VOCs

In order to find the relationship between the deposition doses of VOCs and the environmental concentrations, nine kinds of VOCs including CH_2O, CH_5N, C_2H_6O, $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8, C_8H_8 and C_8H_{10}, were selected (Table S2). These compounds can better illustrate the different features critical for the clinical breath-gas analysis, such as physiological production, environmental exposure, reliance on smoking habits and health (An et al., 2014; Wilson, 2015).

The concentrations of acetone and isoprene in the exhalations of the selected volunteers were clearly increased in the expiratory phase (Fig. S3). This is because acetone and isoprene are the endogenous compounds mainly exhaled during the blood–air exchange in the alveoli (Miekisch et al., 2004). In contrast, the concentrations of benzene decreased from the inhaled concentration of 1.50 ppb to the exhaled concentration of 0.50 ppb, indicating that the benzene is an exogenous substance. Furthermore, the concentration gap between the inhaled and exhaled inspiratory phase was clearly higher than those of the exogenous substances. Therefore, in the present study, acetone and isoprene were selected as the breath tracers to identify the expiratory and inspiratory phases similar to the conclusions presented in the previous studies (Herbig et al., 2009; Huang et al., 2016).

Herein, all the subjects were exposed to different concentrations of VOCs in the same normal laboratory due to diurnal variations. The exhaled concentrations shown in Fig. S3 were the mean concentrations calculated in the exhaled plateau stage. The regression analysis between the deposition concentrations (C_i-C_e) and environmental concentrations (C_i) of 120 healthy volunteers, calculated by Eq. (2), are shown in Fig. 1.

$$C_i\text{-}C_e = C_0 + aC_i \quad (2)$$

In the regression correlation, the intercept C_0 indicates the endogenous concentration of the particular compound when the inhalation concentration is 0. The slope of the linear regression line, $a = (C_i\text{-}C_e\text{-}C_0)/C_i$, can be regarded as the respiratory deposition rate of the test group (Spaněl et al., 2013).

In present study, the exposure concentrations of the VOCs were measured within the range of 1.81–18.42 ppb for CH_2O, 3.20–6.68 ppb for CH_5N, 7.72–88.99 ppb for $C_2H_4O_2$, 5.05–57.86 ppb for C_3H_8O, 0.38–4.13 ppb for C_6H_6, 0.68–12.50 ppb for C_7H_8, 0.14–1.59 ppb for C_8H_8, and 0.45–10.39 ppb for C_8H_{10}. According to the National Institute of Occupational Safety and Health, the concentrations of all the selected VOCs were clearly below the 8-h permissible exposure limits (Spaněl et al., 2013). Therefore, the analyzed VOCs are not detrimental to the human health at present levels. Furthermore, the environmental concentrations of the mentioned VOCs persisted relatively stable. Therefore, the experiments to find the deposition rates of VOCs could be conducted under current the same laboratory conditions. The excessive changes of environmental concentrations are liable to affect the deposition rates of VOCs (Huang et al., 2017). Therefore, the experiments were carried out in the same laboratory conditions to ensure the accuracy of the test. Seven parallel sets of the inhaled and exhaled VOC concentrations of each subject were consecutively measured. The average value of all the seven measured concentrations of each compound was used to plot the regression analysis between the deposition

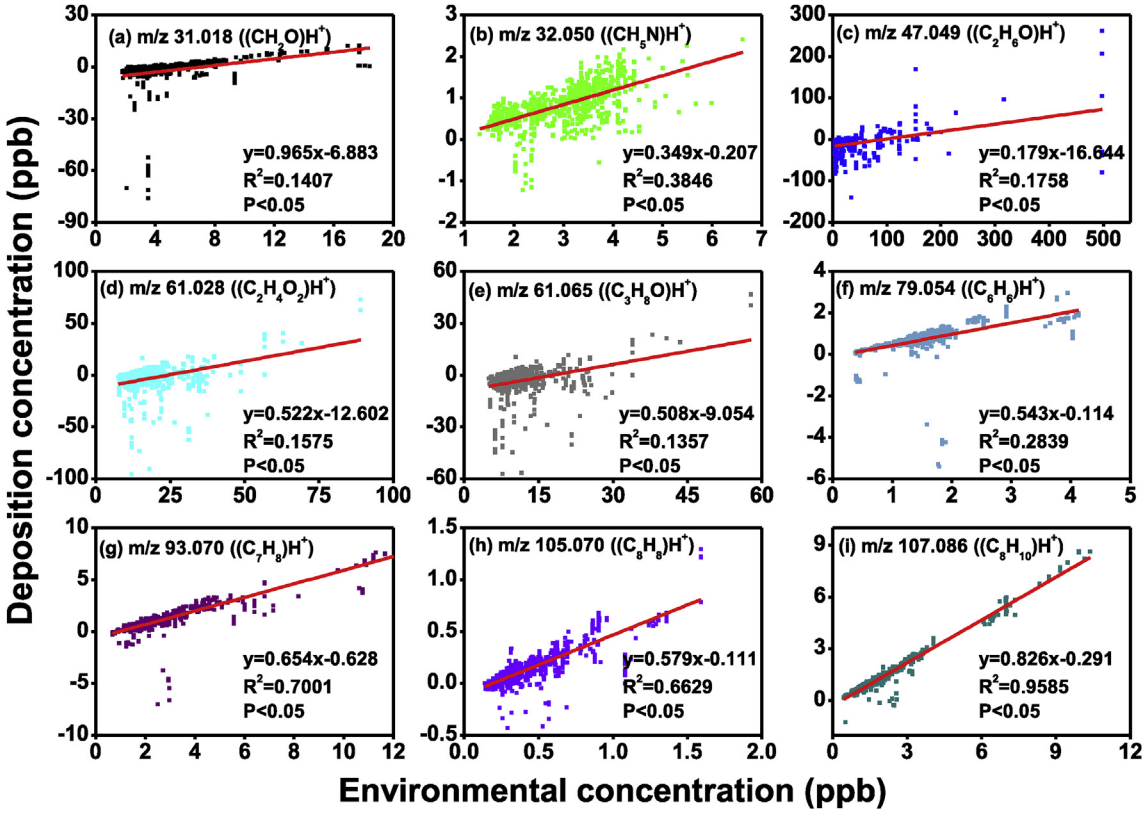

Fig. 1. Plot of regression analysis between deposition and exposure concentrations of selected VOCs (a) CH_2O, (b) CH_5N, (c) C_2H_6O, (d) $C_2H_4O_2$, (e) C_3H_8O, (f) C_6H_6, (g) C_7H_8, (h) C_8H_8 and (i) C_8H_{10} collected from 120 subjects. The inhaled/exhaled concentrations are calculated from PTR-TOF-MS data exemplified in Fig. S2. Curve-fitting equations, correlation coefficients (R^2) and significance levels (P) are presented. $p < 0.05$ were used as critical values for significant correlations.

and exposure concentrations of the selected VOCs. A previously reported method in Eq. (1) (Miekisch et al., 2004) was used to measure the respiratory deposition rates of VOCs.

As shown in Fig. 1, the correlation coefficient (R^2) and significance level ($P < 0.05$) of the linear regression lines suggested that the exogenous VOCs are proportionally deposited in the human respiratory system. These results are consistent with the findings of previous study (Huang et al., 2017). The deposition rates obtained from the regression correlations of CH_2O, CH_5N, C_2H_6O, $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8, C_8H_8 and C_8H_{10} of 120 subjects were 96.6%, 34.9%, 17.9%, 52.2%, 50.8%, 54.3%, 65.4%, 60.9% and 55.5%, respectively, and are shown in Fig. 2 of the red line. The results of deposition rates are more or less similar to the mean respiratory deposition rates of 55.0%, 55.9%, and 66.9% for C_6H_6, C_7H_8, and C_8H_{10}, respectively, as reported in the study conducted by Perbellini et al. who monitored the respiratory deposition rates of some VOCs in the ambient air using PTR-TOF-MS in the surroundings of chemical workers (Perbellini et al., 1988). Therefore, it would appear from our results that, in all the analyzed VOCs, CH_2O has the highest exposure risk, followed by C_7H_8 and C_8H_8, while the C_2H_6O has the lowest exposure risk if the people are exposed to the same concentration of these VOCs via inhalation.

In the present study, the deposition rate of an individual VOC was also calculated from the inhalation and exhalation concentrations of each group according to the method used in Eq. (1). The results obtained were represented by box diagrams as shown in Fig. 2. According to Fig. 2, the analyzed VOCs' deposition rates of regression analysis were higher than the median of the calculated deposition rates except for C_8H_{10}. The slopes of the regression analysis, $a = (C_i\text{-}C_e\text{-}C_0)/C_i$ for endogenous VOCs, were higher than the calculated deposition rates, $K = (C_i - C_e)/C_i$. These results indicated that, in the real health risk assessment, the real deposition rates, which were the calculation data of the deposition rates, were different from the uniform single standard

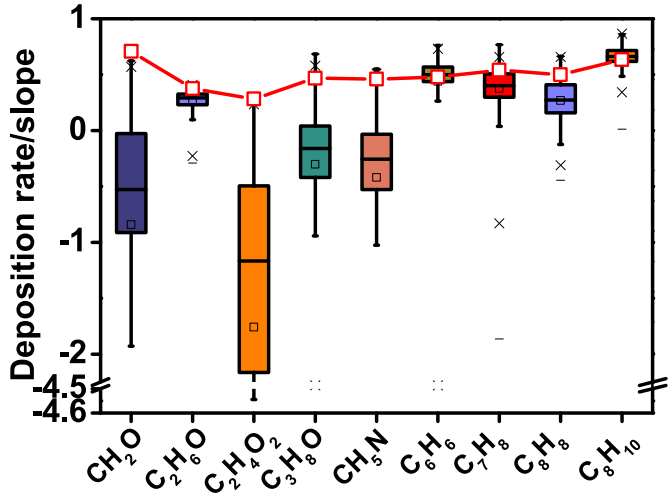

Fig. 2. Box diagram of the deposition rates calculated from the inhaled and exhaled concentration of each set. Line chart of the deposition rates of regression analysis.

deposition rates. Therefore, it is of great significance to calculate the deposition rates of VOCs. In addition, it was evident that the medians of calculated deposition rates of CH_2O and C_2H_6O were less than 0, but the slope deposition rates were greater than 0. This difference may be due to the deposition rate of a single experiment, which was affected by the factors of substance and subjects. The factors influencing the deposition rates of VOCs would be discussed in the following sections.

3.2. Subject factor of influencing the deposition rates of VOCs

When a person inhaled the VOCs present in the environment, the deposition of these compounds in the respiratory system may be different under various conditions. An example is the breathing process determined by the physicochemical properties and the concentrations of VOCs. The physiological parameters as well as the exposure methods of the human body might affect the deposition rates of VOCs. The present study emphasized the effects of deposition rates on the selected VOCs by the factors related to the subjects including the breathing models, circadian variation, age, gender and BMI.

In this experiment, all the subjects were asked to breathe the air present in the laboratory at least 15 times before the experiment to get rid of the pre-exposed inhaled gas. This is because, generally, only one seventh of the exhaled gas in the lungs can be replaced by fresh air, specifically 15 times of breathing can exchange 90.1% of gas in human lungs (Yu et al., 2018). In addition, the inhaled VOCs are predominantly deposited in the fluid of the respiratory tract (Medinsky and Bond, 2001). According to the Fick's law, when the ambient VOC concentrations in the laboratory are high and sufficient, the VOC molecules diffuse from alveolus into the capillary of respiratory system dictated by the partial pressure gradient. A portion of the inhaled VOCs deposits in the humans, while the remaining is exhaled. Therefore, the exhaled concentrations of VOCs are less than the environmental concentrations. As shown in Fig. 1, the inhaled VOCs from the environment were deposited in the human respiratory system, and the deposition rates of VOCs were positive. For example, the deposition rate of C_6H_6 was 54.3%, indicating that approximately 54.3% of C_6H_6 from the environment was deposited in the human respiratory system. Conversely speaking, the VOCs produced in the human body are also excreted through the breathing when the external concentration of VOC is relatively low, demonstrated as a negative value of the deposition rate according to the reference (He et al., 2019). In addition, molecular weight, solubility and lipid-water partition coefficient of the VOCs could also affect their deposition rates. In the present study, the highest deposition rate was found for formaldehyde (Fig. 2), which may possibly be due to its high water solubility, making its deposition in the upper respiratory tract (Spaněl et al., 2013).

3.2.1. The breathing models and circadian variation

The first designed experiment conducted on two males and two females elucidates whether the breathing model difference and the circadian variation, which could affect the respiratory deposition rates of VOCs. As shown in Fig. 3a, the deposition rates of $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8 and C_8H_{10} by two breathing models of nasal-in-oral-out and oral-in-oral-out were more significantly higher than the nasal-in-nasal-out. However, there was no significant difference between the nasal-in-oral-out and oral-in-oral-out models. The possible reason for the difference of the two results is that the oral cavity exposure is mainly affected by the exhaled VOC concentrations (Sukul et al., 2017a). Therefore, the human beings should retain the natural breathing model of nasal-in-nasal-out in case that the VOCs are susceptible to settle in the body. In addition, the water vapor of oral cavity is higher than that of the nasal cavity. As such, the VOCs could be more easily remained in oral cavity, which increases the respiratory health effects. Furthermore, in the present study, from the obtained deposition rates of CH_2O, CH_5N, $C_2H_4O_2$, C_3H_8O and C_6H_6, it can be concluded that the breathing model of oral-in-oral-out is the most effective way to deposit the VOCs in the human respiratory system among all the three breathing models. The deposition rates of C_2H_6O, C_8H_8 and C_8H_{10}, through the nasal-in-oral-out model were higher than those of the other two breathing models. Although there was no significant difference between the breathing models, nasal-in-oral-out and oral-in-oral-out, the nasal-in-oral-out is certainly a breathing model able to expose the whole respiratory system to the VOCs, which was more suitable to evaluate the health risk assessment of respiratory exposure. Therefore, the breathing model of nasal-in-oral-out was selected for the following experiments in the present study.

Moreover, the respiratory deposition rates of VOCs at different circadian variations, including before and after the breakfast, lunch and dinner were also evaluated. As shown in Fig. 3b, no significant difference in the deposition rates of VOCs was obtained at different sampling times related to circadian variation. These results indicate that the circadian variation related to the efficiency of metabolism is not the main factor affecting the deposition rates (Martinez-Lozano Sinues et al., 2014). Therefore, there is no need to limit the sampling time in the later experiments.

3.2.2. Gender, age and BMI

Generally, gender, age and BMI determine the physiological structure and metabolism efficiency. Therefore, it is possible that they may affect the respiratory deposition rates of VOCs via inhalation. In this study, in order to explore whether or not gender, age and BMI of the individuals affect the respiratory deposition rates, the deposition rates of VOCs were measured for 120 subjects. Fig. 3 indicates that the gender and BMI significantly influenced the deposition rates of VOCs, while the age did not. Our statistical analysis also confirmed that there is no significant difference between the mean VOC profiles of male and female ($n = 50$ and 70 respectively, $p > 0.05$). As mentioned earlier, gender usually is not a fundamental factor affecting the deposition rates of VOCs. Our results appear to be consistent with the results reported by Huang et al. (2017).

In the present study, the deposition rates of the selected VOCs from 120 healthy subjects were analyzed. For this purpose, all the individuals were divided into two groups according to the subjects' age of more than 18 years. As shown in Fig. 4, CH_2O had a significantly higher value, while CH_5N and C_2H_6O had significantly lower deposition rates in the young individuals as compared to the individuals of age above 18 years. For remaining 7 types of VOCs, there was no significant difference between the deposition rates of the two groups. These results indicate that the exposure risks of the same VOCs varied significantly in different age groups. Further research is needed to find the reason behind the diverse deposition rates of CH_2O, CH_5N, and C_2H_6O in different age groups.

The World Health Organization (WHO) introduced the BMI as a universal standard to assess the body fat levels (https://www.who.int/). In the present study, in order to find the effect of body fat levels on the deposition rates of VOCs, the measurements available were divided into three groups as follows: low (BMI below 18.5, $n = 20$), normal (BMI 18.5–25, $n = 63$) and obese (BMI over 25, $n = 12$). As shown in Fig. 4, significantly higher deposition rates of CH_2O and CH_5N were detected in the normal groups ($p < 0.05$) as compared with the other two groups. However, there was no significant correlation between the BMI and the deposition rates of C_6H_6, C_7H_8 and C_8H_{10}, which are consistent with the research conducted by Huang et al. (2017). In their study, they showed that the adipose tissue existing within the airway wall is related to BMI. Therefore, the accumulation of airway adipose tissue in obese individuals may occupy the airway space and affect the air exchange functions of the lungs. The deposition of VOCs was susceptible to be hindered in people with high BMI (Elliot et al., 2019). The reason is why low BMI individuals have lower deposition rates than the normal BMI individuals is still unknown. It may be concluded that the reason of BMI influencing the deposition rates of specific VOCs is related to the physical and chemical properties of the specific VOCs.

In short, statistical analysis showed that gender, age and BMI did not significantly affect the deposition rates of nine kinds of investigated VOCs. However, age does affect the deposition rates of CH_2O, CH_5N and C_2H_6O.

3.2.3. Lung function parameters

Pulmonary function parameters are also important indicators for evaluating the lung health of humans. The respiratory function factors

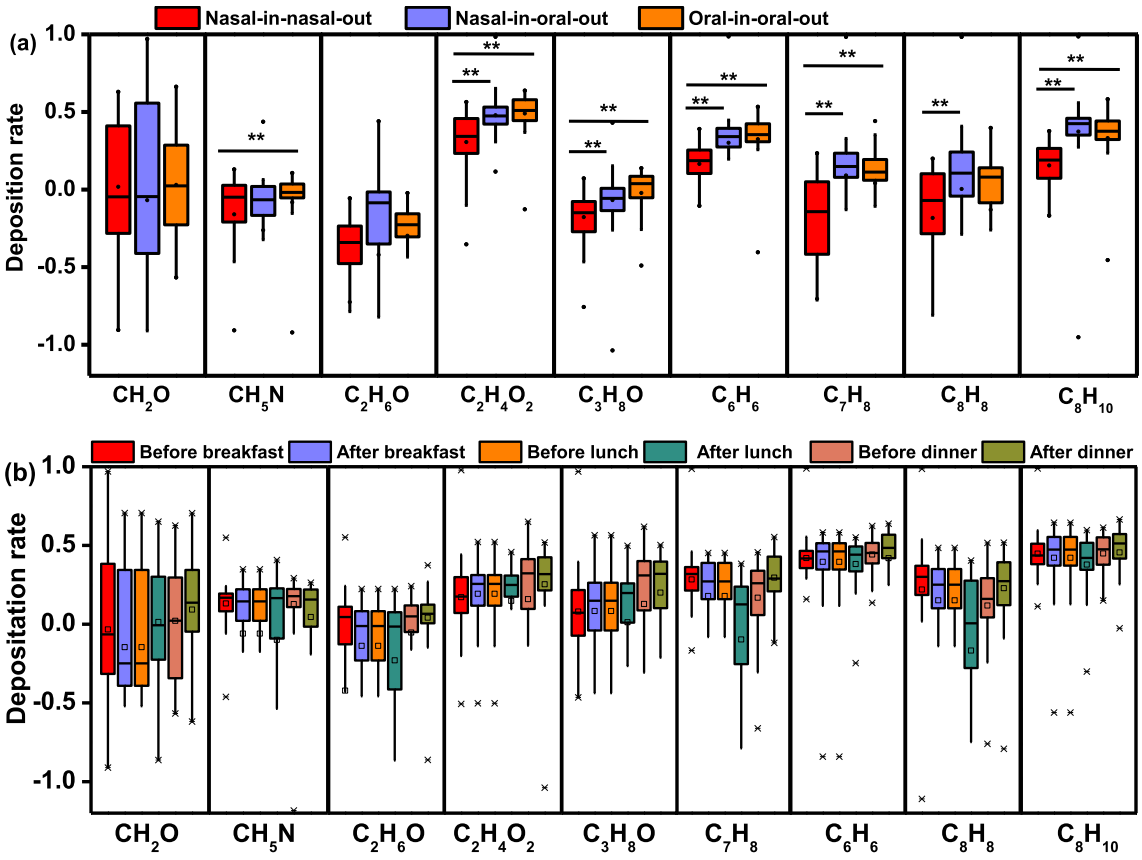

Fig. 3. Subjects adopting self-made human exhaled gas collecting device by (a) three breathing models: nasal-in-nasal-out, nasal-in-oral-out, and oral-in-oral-out were measured deposition rates of 9 kinds of VOCs at (b) 6 sampling time points before and after breakfast, lunch and dinner.

mainly include FEV1, FVC, MMEF, MVV, and VT (tidal volume). Common spirometric indicators of respiratory obstruction disease are FVC, FEV1 and MMEF, while MVV and VT represent lung ventilation. The MMEF is in the non-force part of FVC, whereas the flow is affected by the small airway diameter and the decline in the flow rate reflects the small airway obstruction. Therefore, the lung function parameters can reflect the amount of lung ventilation and reserve of the subjects (Miller et al., 2005).

As shown by the results presented in Table 1, the lung function factors are correlated with each other. The Spearman's correlation coefficients between the lung function factors and the deposition rates of CH_2O, CH_5N, C_6H_6 and C_8H_{10} were within the range of 0.2–0.4, indicating that the correlation between the lung function factors and the deposition rates of the above mentioned four types of VOCs were not very high. Therefore, the lung function factors are not the major aspects influencing the deposition rates of the VOCs in healthy individuals. It is because the area of the respiratory membrane is significantly higher than the actual gas exchange needed (Qureshi and Mustafa, 2018). Due to the least significant difference of the deposition rates, it would appear that there were no pathological changes of lung function in healthy individuals to disturb the deposition rates of the VOCs. On the other hand, there was no relationship between the lung function factors and the deposition rates of C_2H_4O, C_3H_8O, C_6H_6 C_7H_8 and C_8H_8. These results suggested that the deposition rates of these VOCs are not affected by the lung function factors of the individuals. The two opposite results revealed that the deposition rates of not all the VOCs were affected by the lung function factors, but depended on the physicochemical properties of the VOCs (Medinsky and Bond, 2001; Nielsen et al., 2008). The positive relationship between the lung function factors and the deposition rates of VOCs reflected that the larger the lung function parameters, the higher were the deposition rates of the selected VOCs. These results indicated that VOCs were more prone to deposits in the proper ventilated lungs. Overall, we could conclude that the respiratory function are fundamental to influence the deposition rates of CH_2O, CH_5N, C_2H_6O and C_8H_{10}.

4. Conclusions

In order to understand whether or not the physiological factors of humans determine the variability of the respiration deposition rates as well as the potential exposure risk of the nine selected VOCs to the individuals via inhalation, many healthy volunteers were invited to participate in two sets of experiments. A significant difference was observed in individual variability in three breathing models, including nasal-in-nasal-out, nasal-in-oral-out and oral-in-oral-out. The subjects with oral respiration could achieve higher deposition rates of $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8 and C_8H_{10}, which was due to higher oral temperature and humidity. The deposition rate changes of circadian variation were not statistically significant. In addition, no significant difference of the deposition rates of the analyzed VOCs was found in gender, which was different from the BMI and age on the CH_2O, CH_5N and C_2H_6O. The lung function factors impacting the deposition rates of VOCs elucidated that well-ventilated and higher capacity lung were more vulnerable to absorb higher levels of VOCs. The present study statistically proves that the respiratory deposition rates of the VOCs are greatly influenced by the physiological factors. Therefore, the accurate deposition rates are important for properly assessing the inhalation exposure risk.

CRediT authorship contribution statement

Yi Yang: Methodology, Writing - original draft. **Hao Luo:**

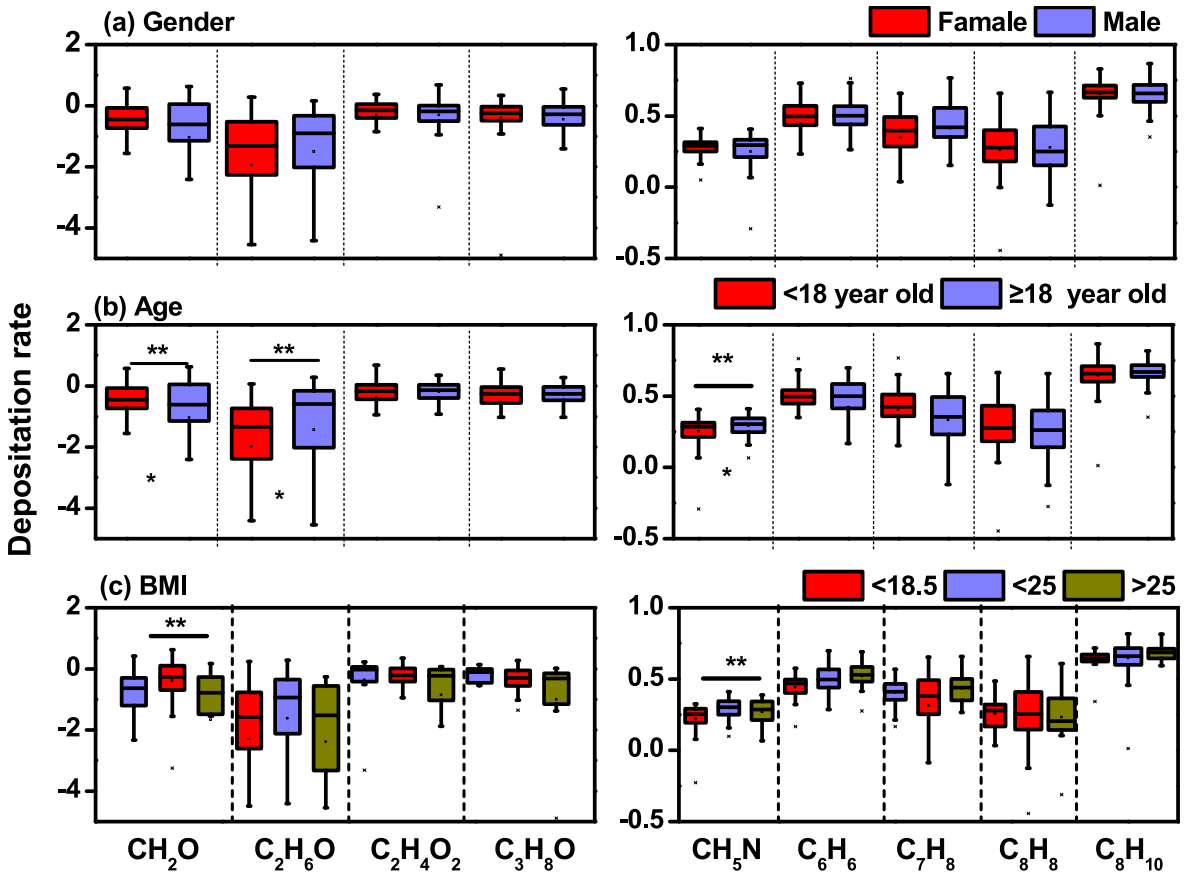

Fig. 4. Box plots of respiratory deposition rates of CH_2O, CH_5N, C_2H_6O, $C_2H_4O_2$, C_3H_8O, C_6H_6, C_7H_8, C_8H_8, and C_8H_{10} in (a) gender, (b) age and (c) BMI collected from 120 subjects.

Table 1
Results from the Spearman correlation analysis.

Factor	FEV1	FVC	MMEF	MVV	VT	CH_2O	CH_5N	C_2H_6O	$C_2H_4O_2$	C_3H_8O	C_6H_6	C_7H_8	C_8H_8	C_8H_{10}
	(L)	(L)	(L/S)	(L/m)	(L)									
FEV1	1	0.962**	0.845**	0.738**	0.685**	0.356**	0.302**	0.249**	−0.043	−0.018	0.086	0.027	0.103	0.205
FVC	0.962**	1	0.709**	0.689**	0.634**	0.285**	0.304**	0.233**	−0.072	−0.055	0.056	0.002	0.077	0.185
MMEF	0.845**	0.709**	1	0.705**	0.668**	0.392**	0.321**	0.185**	0.019	0.037	0.189	0.08	0.135	0.213*
MVV	0.738**	0.689**	0.705**	1	0.866**	0.261*	0.243*	0.156	0.035	0.052	0.084	0.055	0.087	0.219*
VT	0.685**	0.634**	0.668**	0.866	1	0.316**	0.161	0.129	0.009	0.024	0.149	0.092	0.069	0.205

* : $p < 0.05$; ** : $p < 0.001$.
Columns 2–11 show the Pearson relationship coefficients between physiological parameters and deposition rates. Changing from green to red indicates that the correlation coefficient is getting smaller. 0.05 and 0.01 significance levels are indicated by * and **, respectively.
FEV1: Forced expiratory volume in 1 s. FVC: forced vital capacity. MMEF: Maximum mid expiratory flow rate. MVV: maximal ventilator volume. VT: tidal volume.

Validation. **Ranran Liu:** Data curation. **Guiying Li:** Supervision, Writing - review & editing. **Yingxin Yu:** Supervision, Validation. **Taicheng An:** Conceptualization, Supervision.

Declaration of competing interest

The authors declare that they have no conflict of interest.

Acknowledgements

This work was supported by National Natural Science Foundation of China (41731279, 41991310 and U1901210), Local Innovative and Research Teams Project of Guangdong Pearl River Talents Program (2017BT01Z032), The Innovation Team Project of Guangdong Provincial Department of Education, China (2017KCXTD012) and Leading Scientific, Technical and Innovation Talents of Guangdong special support program (2016TX03Z094). The authors also would like to express their sincere thanks to volunteers in Guangdong University of Technology for their support.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecoenv.2020.110615.

References

An, T., Huang, Y., Li, G., He, Z., Chen, J., Zhang, C., 2014. Pollution profiles and health risk assessment of VOCs emitted during e-waste dismantling processes associated with different dismantling methods. Environ. Int. 73, 186–194.
Audi, C., Baiz, N., Maesano, C.N., Ramousse, O., Reboulleau, D., Magnan, A., Caillaud, D.,

Annesi-Maesano, I., 2017. Serum cytokine levels related to exposure to volatile organic compounds and PM2.5 in dwellings and workplaces in French farmers - a mechanism to explain nonsmoking COPD. Int. J. Chronic Obstr. 12, 1363–1374.
Beauchamp, J., 2011. Inhaled today, not gone tomorrow: pharmacokinetics and environmental exposure of volatiles in exhaled breath. J. Breath Res. 5 (3), 037103.
Bikov, A., Paschalaki, K., Logan-Sinclair, R., Horváth, I., Kharitonov, S.A., Barnes, P.J., Usmani, O.S., Paredi, P., 2013. Standardised exhaled breath collection for the measurement of exhaled volatile organic compounds by proton transfer reaction mass spectrometry. BMC Pulm. Med. 13 (1), 43.
Cao, F., Qin, P., Lu, S., He, Q., Wu, F., Sun, H., Wang, L., Li, L., 2018. Measurement of volatile organic compounds and associated risk assessments through ingestion and dermal routes in Dongjiang Lake, China. Ecotoxicol. Environ. Saf. 165, 645–653.
Elliot, J.G., Donovan, G.M., Wang, K.C.W., Green, F.H.Y., James, A.L., Noble, P.B., 2019. Fatty airways: implications for obstructive disease. Eur. Respir. J. 54 (6), 1900857.
Han, C., Liu, R., Luo, H., Li, G., Ma, S., Chen, J., An, T., 2019. Pollution profiles of volatile organic compounds from different urban functional areas in Guangzhou China based on GC/MS and PTR-TOF-MS: atmospheric environmental implications. Atmos. Environ. 214, 116843.
He, J., Sun, X., Yang, X., 2019. Human respiratory system as sink for volatile organic compounds: evidence from field measurements. Indoor Air 29 (6), 968–978.
Herbig, J., Muller, M., Schallhart, S., Titzmann, T., Graus, M., Hansel, A., 2009. On-line breath analysis with PTR-TOF. J. Breath Res. 3 (2), 027004.
Huang, Z., Zhang, Y., Yan, Q., Wang, Z., Zhang, Z., Wang, X., 2017. Decreased human respiratory absorption factors of aromatic hydrocarbons at lower exposure levels: the dual effect in reducing ambient air toxics. Environ. Sci. Technol. Lett. 4 (11), 463–469.
Huang, Z., Zhang, Y., Yan, Q., Zhang, Z., Wang, X., 2016. Real-time monitoring of respiratory absorption factors of volatile organic compounds in ambient air by proton transfer reaction time-of-flight mass spectrometry. J. Hazard Mater. 320, 547–555.
Jakubowski, M., Czerczak, S., 2009. Calculating the retention of volatile organic compounds in the lung on the basis of their physicochemical properties. Environ. Toxicol. Pharmacol. 28 (2), 311–315.
Lewtas, J., 2007. Air pollution combustion emissions: characterization of causative agents and mechanisms associated with cancer, reproductive, and cardiovascular effects. Mutat. Res. 636 (1–3), 95–133.
Li, G., Liao, Y., Hu, J., Lu, L., Zhang, Y., Li, B., An, T., 2019. Activation of NF-κB pathways mediating the inflammation and pulmonary diseases associated with atmospheric methylamine exposure. Environ. Pollut. 252, 1216–1224.
Li, G., Sun, H., Zhang, Z., An, T., Hu, J., 2013. Distribution profile, health risk and elimination of model atmospheric SVOCs associated with a typical municipal garbage compressing station in Guangzhou, South China. Atmos. Environ. 76, 173–180 0.
Liang, Z., Wang, J., Zhang, Y., Han, C., Ma, S., Chen, J., Li, G., An, T., 2020. Removal of volatile organic compounds (VOCs) emitted from a textile dyeing wastewater treatment plant and the attenuation of respiratory health risks using a pilot-scale biofilter. J. Clean. Prod. 253, 120019.
Lindinger, W., Hansel, A., Jordan, A., 1998. Proton-transfer-reaction mass spectrometry (PTR-MS): on-line monitoring of volatile organic compounds at pptv levels. Chem. Soc. Rev. 27 (5), 347–354.
Martinez-Lozano Sinues, P., Tarokh, L., Li, X., Kohler, M., Brown, S.A., Zenobi, R., Dallmann, R., 2014. Circadian variation of the human metabolome captured by real-time breath analysis. PloS One 9 (12), e114422.
Medinsky, M.A., Bond, J.A., 2001. Sites and mechanisms for uptake of gases and vapors in the respiratory tract. Toxicology 160 (1–3), 165–172.
Miekisch, W., Schubert, J.K., Noeldge-Schomburg, G.F.E., 2004. Diagnostic potential of breath analysis—focus on volatile organic compounds. Clin. Chim. Acta 347 (1–2), 25–39.
Miller, M.R., Hankinson, J., Brusasco, V., Burgos, F., Casaburi, R., Coates, A., Crapo, R., Enright, P., van der Grinten, C.P., Gustafsson, P., Jensen, R., Johnson, D.C., MacIntyre, N., McKay, R., Navajas, D., Pedersen, O.F., Pellegrino, R., Viegi, G., Wanger, J., Force, A.E.T., 2005. Standardisation of spirometry. Eur. Respir. J. 26 (2), 319–338.
Nielsen, E., Østergaard, G., Larsen, J.C., 2008. Toxicological Risk Assessment of Chemicals. A Practical Guide.
Omidi, F., Dehghani, F., Fallahzadeh, R.A., Miri, M., Taghavi, M., Eynipour, A., 2019. Probabilistic risk assessment of occupational exposure to volatile organic compounds in the rendering plant of a poultry slaughterhouse. Ecotoxicol. Environ. Saf. 176, 132–136.
Perbellini, L., Faccini, G.B., Pasini, F., Cazzoli, F., Pistoia, S., Rosellini, R., Valsecchi, M., Brugnone, F., 1988. Environmental and occupational exposure to benzene by analysis of breath and blood. Br. J. Ind. Med. 45 (5), 345–352.
Qureshi, S.M., Mustafa, R., 2018. Measurement of respiratory function: gas exchange and its clinical applications. Anaesth. Intens. Care 19 (2), 65–71.
Rumchev, K., Spickett, J., Bulsara, M., Phillips, M., Stick, S., 2004. Association of domestic exposure to volatile organic compounds with asthma in young children. Thorax 59 (9), 746–751.
Spaněl, P., Dryahina, K., Smith, D., 2013. A quantitative study of the influence of inhaled compounds on their concentrations in exhaled breath. J. Breath Res. 7 (1), 017106.
Sukul, P., Oertel, P., Kamysek, S., Trefz, P., 2017a. Oral or nasal breathing? Real-time effects of switching sampling route onto exhaled VOC concentrations. J. Breath Res. 11 (2), 027101.
Sukul, P., Schubert, J.K., Kamysek, S., Trefz, P., Miekisch, W., 2017b. Applied upper-airway resistance instantly affects breath components: a unique insight into pulmonary medicine. J. Breath Res. 11 (4), 047108.
Sukul, P., Schubert, J.K., Oertel, P., Kamysek, S., Taunk, K., Trefz, P., Miekisch, W., 2016. FEV manoeuvre induced changes in breath VOC compositions: an unconventional view on lung function tests. Sci. Rep. 6, 28029.
Tagiyeva, N., Sheikh, A., 2014. Domestic exposure to volatile organic compounds in relation to asthma and allergy in children and adults. Expet Rev. Clin. Immunol. 10 (12), 1611–1639.
Wei, W., Bonvallot, N., Gustafsson, A., Raffy, G., Glorennec, P., Krais, A., Ramalho, O., Le Bot, B., Mandin, C., 2018. Bioaccessibility and bioavailability of environmental semi-volatile organic compounds via inhalation: a review of methods and models. Environ. Int. 113, 202–213.
Weisel, C.P., Jo, W.K., 1996. Ingestion, inhalation, and dermal exposures to chloroform and trichloroethene from tap water. Environ. Health Perspect. 104 (1), 48–51.
Wilson, A.D., 2015. Advances in electronic-nose technologies for the detection of volatile biomarker metabolites in the human breath. Metabolites 5 (1), 140–163.
Xu, X.H., Freeman, N.C., Dailey, A.B., Ilacqua, V.A., Kearney, G.D., Talbott, E.O., 2009. Association between exposure to alkylbenzenes and cardiovascular disease among national health and nutrition examination survey (NHANES) participants. Int. J. Occup. Environ. Health 15 (4), 385–391.
Yang, K., Wang, C., Xue, S., Li, W., Liu, J., Li, L., 2019. The identification, health risks and olfactory effects assessment of VOCs released from the wastewater storage tank in a pesticide plant. Ecotoxicol. Environ. Saf. 184, 109665.
Yu, Y., Jiang, Z., Zhao, Z., Chong, D., Li, G., Ma, S., Zhang, Y., An, T., 2018. Novel in vitro method for measuring the mass fraction of bioaccessible atmospheric polycyclic aromatic hydrocarbons using simulated human lung fluids. Environ. Pollut. 242 (Pt B), 1633–1641.
Yuan, B., Koss, A.R., Warneke, C., Coggon, M., Sekimoto, K., de Gouw, J.A., 2017. Proton-transfer-reaction mass spectrometry: applications in atmospheric sciences. Chem. Rev. 117 (21), 13187–13229.