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The exposures and health effects of benzene, toluene and naphthalene for Chinese chefs in multiple cooking styles of kitchens

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A R T I C L E I N F O

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A B S T R A C T

Commercial cooking has higher intensity and more severe instantaneous cooking pollution from volatile organic chemicals compared to home cooking, making health risk assessment of occupational exposure for chefs a priority. In this study, chefs from three cooking styles of kitchens, including steaming, frying, and grilling, were selected to investigate the external and internal exposures, health risks and effects of several typical aromatic hydrocarbons (benzene, toluene and naphthalene). Naphthalene was found to be the most concentrated contaminant in air samples among the different kitchens, while benzene had the lowest concentration. The concentration of toluene in frying kitchens was significantly higher than that in steaming kitchens. Air concentrations of toluene in frying kitchens, as well as benzene concentrations in grilling kitchens exceeded the standard level according to indoor air quality standard (GB/T18883-2002). Regarding the metabolites of pollutants in urine, the content of S-benzylmercapturic acids (S-BMA) for frying chefs was significantly higher than that for other cooking styles of chefs, which was consistent with the relatively higher air concentrations of toluene. There was a good correlation between internal and external exposure of the pollutants. The level of oxidative stress was influenced by 2-hydroxynaphthalene (2-OHN) and S-BMA, indicating the potential health risks of these occupational exposed chefs. This study indicates the need to improve the monitoring of typical aromatic hydrocarbons, as well as to investigate their potential health effects in large-scale groups, and improve the ventilation in kitchens.

1. Introduction

From 1990 to 2017, the number of premature deaths caused by household air pollution associated with solid fuel use dropped by 39% worldwide, while scholars are paying more attention to the health effects of fine particles (Orach et al., 2021; Yang et al., 2020). However, the type of cooking fuel has more significant impact on the risk reduction of premature death (Zhao et al., 2018). Globally, nearly three billion people still cook with solid fuels or kerosene in open fires with inefficient stoves. Such inefficient burning process and fuel-using technologies have caused severe indoor air pollution, which could lead to 3.8 million deaths per year (Health Effects Institute, 2019). Household air pollution is gradually receiving research attention. In particular, cooking contributes significantly to the total $PM_{2.5}$ from residential emissions in indoor air (Yun et al., 2020).

The rapid development of catering industry is highly beneficial to the prosperity of China's economy. However, cooking emission is one of the major contributors to indoor air pollution (Chen et al., 2018; Lu et al., 2020). The cooking process produces many pollutants, including numerous gases and inhalable particles. Cooking pollutants can be an important source of volatile organic compounds (VOCs) including carbonyl compounds, as well as particulates (Alves et al., 2015; Nayek and Padhy, 2020). China first introduced a regulation in 2002 to prevent the pollution of atmospheric and residential environments caused by cooking fumes from the catering industry, which was revised in 2019 to include kitchen emission VOCs into the monitoring lists. It has been

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reported that the style of cooking, especially deep-frying/stir-frying cooking in iron pan with unrefined rapeseed oil, led to massive indoor air pollutant emission and increased the risk of lung cancer in Shanghai, China (Zhong et al., 1999). There has been an epidemiological association of cooking with lung adenocarcinoma in nonsmoking women (Mu et al., 2013). Besides, the risks of oxidative stress, respiratory tract infection, lung disease, heart disease, stroke, eye disease, tuberculosis and cancer are higher in Chinese chefs. Some of these diseases have been included in the occupation disease classification and catalogue in China (NHFPC, 2013). Now, clean fuel may reduce the harmful health effects associated with household air pollution and even reduce the prevalence of low birth weight (Alexander et al., 2017; Imelda, 2020). Whether other cooking factors will have different health effects is worth figuring out.

Commercial cooking is more intense, lasts longer and produces higher concentrations of instantaneous cooking contaminants than home cooking, and the resultant impact on human health is of greater concern (Franklin et al., 2006). The amount and composition of pollutants emitted from cooking largely depend on what kind of food is cooked, and how it is cooked (Kabir and Kim, 2011). It can also be considered that different cooking methods have different cooking pollutant components due to different types of cooking oil, materials, temperature, and time control. Different cooking methods in the kitchen would lead to different levels of indoor air pollution. Steaming and frying are the most common cooking methods in the world (Lim et al., 2019; Sun et al., 2020). Grilling is one of the oldest cooking methods still used today and is highly appreciated by consumers all over the world (Echegaray et al., 2020; He, 2012; Wu et al., 2015). Among the common cooking methods, deep-frying and stir-frying produce more polycyclic aromatic hydrocarbons (PAHs) than other cooking methods with less oil (Abdullahi et al., 2013). Stir-frying and grilling tend to produce gaseous pollutants (Sinha and Ray, 2015). Compared with occupational safety accidents leading to mass death and mass injury, occupational diseases are often neglected by the managers and practitioners themselves.

Thus, it is very important to study the kitchen air pollutants and the resultant occupational health effects from commercial cooking on different styles of chefs. This study focuses on the emission and health effects of typical aromatic hydrocarbon pollutants from cooking, such as benzene, toluene, naphthalene as well as their metabolites. Benzene is listed as a confirmed human carcinogen by the International Agency for Research on Cancer (IARC) under the World Health Organization, while naphthalene is identified as possibly carcinogenic to humans, and toluene can cause harm to the nervous system (Amor-Carro et al., 2020; Kampa and Castanas, 2008). However, research on this subject is very limited as far as we know. To better understand the relationship between benzene, toluene and naphthalene from cooking and the resulting health impacts on chefs, three kinds of kitchens were selected in Nanjing, China for the research with Chinese chefs. First, the external exposure of benzene, toluene and naphthalene was evaluated by monitoring the main pathway of inhalation. Second, the internal exposure of three typical aromatic hydrocarbons was analyzed by testing urinary concentrations of their metabolites. Third, the relationship between internal and external exposure levels was evaluated. Finally, the health effects of the three typical aromatic hydrocarbons were studied, including oxidative stress, lung function, and blood pressure.

2. Material and methods

2.1. Study subjects and sample collection

For this panel study, chefs from steaming, frying, and grilling kitchens were selected and surveyed. The questionnaire elicited basic personal information such as the subject's age, height, weight, smoking status and exposure time to kitchens during the previous 24 h, as well as days of exposure per year (i.e., exposure frequency). The study was approved by the Special Committee of Academic Ethics and Technology Ethics of Guangdong University of Technology. Consent was obtained from all participants after the purpose of this study was clearly stated. Morning urine, saliva, and oral epithelial cell samples were collected from six chefs aged 18 – 55 in each style of steaming, frying, and grilling kitchens, respectively. The samples were frozen and stored at –80 °C until analysis. All the chefs wore monitoring equipment to measure the real-time respiratory rate, systolic blood pressure (SBP), diastolic blood pressure (DBP) and the ratio between forced expiratory volume within 1 s (FEV1) and forced vital capacity (FVC). Both the samples and cardiopulmonary indicators were collected in four seasons in 2017 (that is, in March, June, October and November, respectively).

Before starting the study, it was ensured that the same style kitchen's cooking food types, commercial kitchen size, and passenger flow were kept consistent or similar as far as possible. Three cooking styles of kitchens (steaming, frying, and grilling), and three for each style, were selected from the workplace of the studied chefs. They were all located on the first floor and had similar architectural age and surrounding environments. Field workers for collection of VOCs (benzene, toluene, and naphthalene) visited all studied kitchens during sampling period, and recorded all the relevant information (**Table S1**). Seasonal differences were not shown in the concentration of pollutants in the kitchen (**Table S2**), so VOCs were collected and analyzed only one season rather than every season. Then, aluminum bags (capacity: 2 L) equipped with adjustable flow pump were used to collect VOCs emitted from different cooking processes. The flow rate was set at 50 mL min^{-1}, and the sampler height was kept at about 1.5 m above the ground, in the breathing zones of these chefs. Each kitchen was sampled three times (three working days of a week, eighty minutes each day, no smoking in the kitchen during the sampling).

2.2. Chemicals

Standards of benzene and toluene were obtained from Linde (1 ppm, >99.99%). Naphthalene was obtained from Sigma-Aldrich (Saint Lewis, USA). S-Phenylmercapturic acid (S-PMA) and S-benzylmercapturic acid (S-BMA), as well as their corresponding internal standards S-PMA-d_5 and S-BMA-d_5 were purchased from Toronto Research Chemicals (Toronto, Canada). 1-Hydroxynaphthalene (1-OH-Nap) and its internal standard 2-OH-Nap-d_7 were obtained from Chiron AS (Trondheim, Norway) and Toronto Research Chemicals (Ontario, Canada), respectively. The enzyme β-glucuronidase/arylsulfatase (Helix pomatia) was obtained from Roche (Mannheim, Germany). Methanol and acetonitrile (HPLC grade) were purchased from Merck (Darmstadt, Germany). Acetic acid (HAc), sodium acetate (NaAC) and ammonium acetate (NH_4Ac) were purchased from CNW.

2.3. Sample analysis

The concentrations of benzene, toluene and naphthalene collected in aluminum bags were measured by a gas chromatograph coupled with an ion trap mass spectrometer (Thermo Scientific ITQ1100, USA) within 3 d of sampling according to literature (MEE, 2015).

The concentrations of S-PMA, S-BMA and 2-OHN, which were the main metabolites of benzene, toluene and naphthalene in urine, respectively, were selected as the internal exposure levels of chefs. The analysis of urinary metabolite of naphthalene (2-OHN) was performed based on a previous report (Lin et al., 2020), and the analysis of urinary metabolites of benzene and toluene, S-PMA and S-BMA, was based on an online column-switching liquid chromatography-tandem mass spectrometry method (Schettgen et al., 2008). In brief, one milliliter urine sample was mixed with internal standard (3 ng of S-PMA-d_5 and S-BMA-d_5), and the pH was adjusted to 4 with acetic acid (0.5 M). Then, it was centrifuged for 10 min (12000 r min^{-1}, 4 °C), and the supernatant was taken and placed in an autosampler vial. Further enrichment and separation of the target compounds S-PMA and S-BMA were performed online by column switching between a solid phase extraction (SPE) column (Eclipse Plus C18, 4.6 × 30 mm × 3.5 μm, Agilent Technologies, USA) and a Poroshell 120 EC-C18 (100 mm × 4.6 mm, 2.7 μm, Agilent Technologies, USA) analytical column. The detection of the target was achieved using a liquid chromatograph (Agilent 1260, USA) coupled with tandem mass spectrometer (Agilent 6470, USA). The mobile phase was methanol and water (with 0.1% acetic acid). The full details of the gradient program of the mobile phase and the mass spectra conditions are given in Supporting Information (SI) (**Tables S3-S4**).

Other biological indicators, such as cotinine and 8-OHdG, were detected by the enzyme-linked immunosorbent assay (ELISA) method (Saito et al., 2000; Wang and Song, 2008). MDA was detected by using micro MDA assay kits according to the manufacturer's instructions (JKBio, Shanghai), and creatinine was detected by picric acid spectrophotometry to adjust the level of MDA, 8-OHdG and internal exposure of each individual (MOH, 1996).

2.4. Quality assurance and quality control

Before air sampling, the sampling bag was cleaned with field air for 2 – 3 times. The sampling bag blank and field blank were analyzed by the same procedure as the air samples. The residues of target chemicals in blanks accounted for less than 3% of the kitchen air and were blank-corrected. Kitchen background air samples were also collected during off-working hours. Comparable concentrations of the pollutants were observed among different kitchens and were also background-corrected. For the analysis of urine samples, a matrix spike and a procedure blank were run with every batch of ten samples. The spiked recovery of the blank sample was obtained as 103% – 116% after correction for the internal standard, and the relative standard deviation of the parallel sample was 7% – 16%. Reported concentrations were not surrogate-recovery corrected. The values of method detection limit (MDL), defined as the mean blank mass plus three standard deviations or a signal five times the noise level, were 0.05, 0.3 and 0.4 ng mL^{-1} for 2-OHN, S-PMA and S-BMA, respectively.

2.5. Statistical analysis

All data were analyzed using SPSS (Version 22.0) and R (Version 3.4.1). One-way ANOVA and Kruskal-Wallis test were used to analyze the difference between internal and external exposure levels. Before the difference analysis, normality test was performed on original data of the sample or the data after logarithm conversion. If the normal distribution was satisfied and the variance was homogeneous, one-way ANOVA was used; otherwise, the Kruskal-Wallis test was used. In addition, the health effect regression analysis was performed using the generalized linear mixed-effect model (GLMM). GLMM is suitable for dealing with non-independent and heteroscedastic data in repeated measurements when the reaction variables do not meet the normal distribution. Dependent variables included measured values of certain health indicators at each sampling time for each research subject, such as SBP, DBP, FEV1/FVC, 8-OHdG and MDA. The concentrations of S-PMA, S-BMA and 2-OHN were taken as independent variables in the model, which was controlled by other variables including age, smoking status (i.e., cotinine concentration) and body mass index (BMI), respectively.

2.6. Health risk assessments

Health risk assessments were performed referring to the human health assessment manual (Part F: supplement to inhalation risk assessment guidelines) issued by the United States Environmental Protection Agency (USEPA). In this study, it was assumed that the chefs took one day off each month and had a working life of 40 years (He and Zhang, 2010). The risk of occupational exposure to cancer for each chef was calculated by incremental lifetime cancer risk (ILCR):

$$EC = \frac{CA \times ET \times EF \times ED}{AT} \tag{1}$$

$$CDI = \frac{CA \times IR \times EF \times ED}{BW \times AT} \tag{2}$$

$$HQ = \frac{EC}{RfC} \tag{3}$$

$$ILCR = CDI \times SF \tag{4}$$

Here EC (mg m^{-3}) represents exposure concentration; CA (mg m^{-3}) represents air pollutant concentration (the average concentration of benzene, toluene and naphthalene in each kitchen was calculated); ET (h d^{-1}) represents exposure time; EF (d a^{-1}) represents exposure frequency; ED (a) represents exposed length of working, 40a; AT (d) represents average exposure time, during the carcinogenic assessment, the average lifetime (per capita life expectancy × 365 d a^{-1} × 24 h d^{-1}) was adopted, and during the non-carcinogenic assessment, the average period of exposure cycle (ED × 365 d a^{-1} × 24 h d^{-1}) was adopted; HQ (mg m^{-3}) represents hazard quotient; RfC represents reference concentration of inhalation toxicity; and SF (kg d mg^{-1}) represents carcinogenic slope factor. The calculation parameters are shown in **Table S5**.

3. Results and discussion

3.1. External exposure and risk assessment of benzene, toluene, and naphthalene

Compared with ingestion and dermal exposure, inhalation is the main pathway of external exposure of volatile aromatic hydrocarbons to chefs when cooking in the kitchen. Hence, the inhalation concentration was used as the key index to calculate the external exposure level of chefs. The air concentrations of benzene, toluene, and naphthalene in three cooking styles of kitchens are shown in Fig. 1. The highest concentrations were observed for naphthalene, followed by toluene and benzene. With respect to different kitchens, the air concentration of benzene in the grilling kitchen (129.8 ± 163.2 μg m^{-3}) exceeded the guideline value (0.11 mg m^{-3}) of China's Indoor air quality standard (MOH, 2002) and was significantly higher than the corresponding values of steaming (52.24 ± 55.38 μg m^{-3}) and frying kitchens (71.58 ± 79.39 μg m^{-3}). On the other hand, the air concentration of naphthalene in the frying kitchen (529.0 ± 715.2 μg m^{-3}) was comparable to that in the grilling kitchen (501.2 ± 653.6 μg m^{-3}). They were both slightly higher than the naphthalene concentration in the steaming kitchen (400.8 ± 460.0 μg m^{-3}). However, there were no significant differences for benzene and naphthalene among different kitchens ($p = 0.663$ and $p = 0.961$), suggesting that the three cooking styles had similar

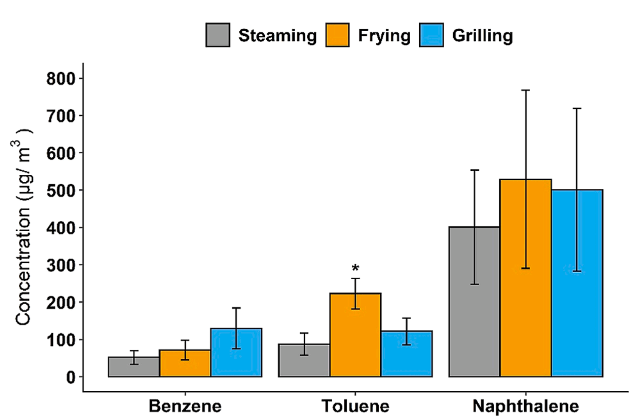

Fig. 1. Air concentrations of benzene, toluene and naphthalene among three different kitchens. (* Compared with steaming kitchen rooms, $p < 0.05$).

production of benzene and naphthalene.

The air concentration of toluene in the frying kitchen (222.6 ± 122.2 μg m^{-3}) was higher than that in the grilling kitchen (122.1 ± 107.0 μg m^{-3}) without a significant difference, but it was significantly higher than that in the steaming kitchen (87.58 ± 87.42 μg m^{-3}, p = 0.013). This may be related to the emission of toluene from other unknown sources. In the present study, natural gas was the main fuel used in the different cooking styles of kitchens, thus the different emission rates caused by fuels is negligible. One explanation could be the use of different types of oils as well as different cooking temperatures in the three styles of kitchens. For instance, heating soybean oil releases higher amount of total VOCs than peanut oil, while heating peanut oil produces higher fraction of benzene (20.8%) compared to soybean oil (10.1%) (Wang et al., 2020). As reported by Schauer et al., the emission rates of aromatic hydrocarbons vary with different grill temperatures used, and higher heating temperatures favor the emission of aromatic hydrocarbons (Schauer et al., 2002). In addition, different seasonings and dishes might impact the emissions of aromatic hydrocarbons (Gao et al., 2019; Wang et al., 2020). The emissions of toluene from single-use kitchen utensils would also make great contribution to the pollutant concentrations detected in different kitchens (Marc, 2020). In addition, studies have shown that the emission of toluene during hot pot, barbecue, and charcoal cooking is significantly greater compared to other cooking methods (Alves et al., 2015; Cheng et al., 2016; Leeu et al., 2001).

The concentration of naphthalene in the present investigation was also obviously lower than that in the indoor air of other commercial kitchens (mean: 3.1 mg m^{-3}) from Lucknow, North India (Singh et al., 2016), but it was higher than that previously reported from commercial and domestic kitchens in Hangzhou, China, with a range of 2.7–9.9 μg m^{-3} (Zhu and Wang, 2003). Nevertheless, these studies indicate that indoor air of kitchen might be polluted by emissions from cooking, which are consistent with our hypothesis.

In addition to external exposure level, the possible health risks of the pollutants also need to be considered. The health risks of benzene, toluene and naphthalene were calculated based on Eq. (4) (Table 1). According to Sexton et al., when the value of ILCR > 1.0×10^{-4}, chemicals are labeled as a definite risk; when 1.0×10^{-5} < ILCR < 1.0×10^{-4}, chemicals are labeled as a probable risk; and when 1.0×10^{-6} < ILCR < 1.0×10^{-5}, chemicals are labeled as a possible risk (Sexton et al., 2007). Benzene was found to be a highly probable risk in the steaming and frying kitchens, while the carcinogenic risk of benzene in the grilling kitchens had a definite risk. The carcinogenic risk of benzene in kitchens should be considered seriously, especially in the grilling kitchens. Naphthalene had a serious non-carcinogenic risk (HQ = 59.66–83.01 > 1) in all kitchens. However, after the US National Toxicology Program revealed evidence of carcinogenic activity of naphthalene in rats, more and more international organizations reexamined the potential carcinogenicity of naphthalene and studied the selection of relevant health parameters (Preuss et al., 2003). Thus, monitoring naphthalene levels in the occupational exposure environment may be one of the key factors for cancer risk assessment in future studies.

3.2. Internal exposure by testing urinary concentrations of metabolites

In order to determine the internal exposure of investigated chefs to benzene, toluene and naphthalene, the urinary concentrations of their main metabolites such as S-PMA, S-BPA and 2-OHN were tested. The basic data of the investigated chefs, such as the urinary cotinine concentration, BMI and age, were used as confounding factors, as shown in Table 2. The statistical results reveal that there were no significant differences in urinary cotinine concentration, BMI and age among different groups of chefs (p = 0.099, p = 0.196, p = 0.533), and the distribution of these three confounding factors of internal exposure among the three kitchen categories was well balanced. Therefore, the confounding factors were excluded from the following category difference analysis.

The urinary concentrations of metabolites, such as S-PMA, S-BPA and 2-OHN, in different cooking styles of chefs are shown in Table 3. The urinary concentration of 2-OHN was higher than that of S-PMA and S-BPA in all styles of chefs. This was also consistent with the relatively higher air concentrations of naphthalene in kitchen rooms. The concentrations of S-PMA and 2-OHN showed no significant differences among the three styles of chefs. Moreover, the median level of S-BMA exposure in frying chefs was 2.25 μmol mol^{-1} Cr, which was significantly higher compared to steaming chefs (1.28 μmol mol^{-1} Cr, p = 0.044) and grilling chefs (0.81 μmol mol^{-1} Cr, p = 0.012). Several studies have investigated the urinary levels of metabolites after exposure to polycyclic aromatic hydrocarbons, benzene, and toluene. S-PMA concentrations ranging from 29 to 95 ng mL^{-1} were found in urine samples collected from kitchen workers (Chauhan et al., 2015), which was higher than the present study (median: 0.0021, 0.092 and 0.029 μmol mol^{-1} Cr). The levels of S-PMA in urine of firefighters (0.28 μmol mol^{-1} Cr) were also considerably higher than chefs in this study (Rosting and Olsen, 2020). On the contrary, the concentrations of S-BMA (2.6 μmol mol^{-1} Cr) reported from the same firefighters were comparable with chefs in this study (median: 1.28, 2.25 and 0.81 μmol mol^{-1} Cr) (Rosting and Olsen, 2020) . The urinary concentration of S-BMA in chefs of the present study was also in the same range as that of occupationally exposed coke plant workers from Southwest China (median: 2.4 μmol mol^{-1} Cr) (Fan et al., 2014). However, the urinary levels of 2-OHN from these coke plant workers (median: 1.2 μmol mol^{-1} Cr) were an order of magnitude lower than chefs in this study (median: 5.79, 8.98 and 9.98 μmol mol^{-1} Cr) (Fan et al., 2014). As reported by Li et al., similar concentrations of 2-OHN (median: 8.5 μmol mol^{-1} Cr) were observed in urine of women tasked with cooking in households using burning wood on indoor open-pit stoves (Li et al., 2011). In addition, the urinary levels

Table 2
Basic information of the chef respondents.

Cooking style	Steaming	Frying	Grilling	p-value [a]
	Mean ± SD	Mean ± SD	Mean ± SD	
Cotinine (μg $mmol^{-1}$)[b]	20.5 ± 16.4	12.0 ± 7.87	13.3 ± 8.38	0.099
BMI (grade)	2.33 ± 1.27	2.25 ± 0.44	2.50 ± 0.78	0.196
Age (year)	31.3 ± 8.27	33.2 ± 7.69	32.6 ± 11.8	0.533

[a] Difference among groups.
[b] The urinary cotinine was generally used as a biomarker of tobacco smoke exposure (Paci et al., 2018).

Table 1
Health risks of benzene, toluene, and naphthalene from different kitchens.[a]

Chemicals	RfC (mg m^{-3})	SF (mg^{-1} kg d)	Hazard Quotient (HQ)			Incremental Lifetime Cancer Risk (ILCR)		
			Steaming	Frying	Grilling	Steaming	Frying	Grilling
Benzene	3×10^{-2}	0.0146	0.83	1.03	2.15	5.53×10^{-5}	7.18×10^{-5}	1.42×10^{-4}
Toluene	5		0.008	0.019	0.012			
Naphthalene	3×10^{-3}		59.66	75.80	83.01			

[a] RfC represents reference concentration of inhalation toxicity; SF represents carcinogenic slope factor; RfC and SF values of target pollutants were obtained from references. (IRIS, 2019; Li et al., 2017).

Table 3
Urinary concentrations of S-PMA, S-BPA and 2-OHN from chefs worked in different styles of kitchens.

Chemicals		Steaming (n = 6)	Frying (n = 6)	Grilling (n = 6)
S-PMA (μmol mol^{-1} Cr)	Mean	0.065 ± 0.11	0.097 ± 0.095	0.13 ± 0.19
	Median	0.0021 (0.00, 0.094)[b]	0.092 (0.0072, 0.16)	0.029 (0.00, 1.21)
	p-value	> 0.05	> 0.05	> 0.05
S-BMA (μmol mol^{-1} Cr)	Mean	1.41 ± 1.19	3.31 ± 3.21	1.56 ± 2.12
	Median	1.28 (0.47, 2.13)	2.25 (0.98, 4.57)	0.81 (0.00, 2.12)
	p-value	> 0.05	**0.044*** [a]	**0.012** #,[a]
2-OHN (μmol mol^{-1} Cr)	Mean	7.33 ± 7.73	9.34 ± 4.71	11.09 ± 7.39
	Median	5.79 (3.36, 9.57)	8.98 (5.55, 12.44)	9.98 (5.37, 17.43)
	p-value	> 0.05	> 0.05	> 0.05

[a] *Compared with steaming chefs, # Compared with frying chefs.
[b] Median (interquartile range).

of 2-OHN were also comparable with gas station workers from Italy (mean: 9.91 ± 9.29 μmol mol^{-1} Cr) (Barbieri et al.). Although reports have indicated the relatively higher urinary concentrations of aromatic hydrocarbon metabolites in occupational exposed chefs, these metabolite levels varied among different groups, which might be derived from various sources in the different styles of kitchens. Different fuels used as well as different ventilation conditions in the kitchen may be the main factors influencing the exposure of the chefs (Li et al., 2011; Zhao et al., 2019). The level of S-BMA exposure in frying chefs was obviously higher than other chefs, which may be due to the high toluene concentration in frying kitchens compared to steaming and grilling kitchens. This result further indicates that the exposure to cooking oil fumes may be the main source of exposure to toluene for frying chefs.

3.3. The relationship between external exposure and internal exposure

External exposure can only affect the human body when exposed inside. In order to understand whether external exposure has an impact on internal exposure, internal and external exposure correlation was conducted. Benzene had a significant positive correlation with S-PMA (r = 0.345, p = 0.004). There was also a significant positive correlation between toluene and S-BMA (r = 0.262, p = 0.031), and naphthalene had a significant positive correlation with 2-OHN (r = 0.252, p = 0.037) (Table 4). Hotz et al. found that S-PMA concentration was a fairly good indicator of benzene exposure in the range of 0.1 to 1 ppm for garage and plant workers, with a correlation coefficient of 0.41 (p = 0.0001) (Hotz et al., 1997), which was similar to the results of this study. Past research showed that urinary S-BMA of factory workers was related to the concentration of toluene in the air at the time of intense exposure. The lowest toluene concentration at which urinary S-BMA rose to measurable levels was approximately 10 ppm, and end-of-shift urinary S-BMA was a good indicator of occupational toluene exposure (Inoue et al., 2002). However, it was not proportional to toluene concentration in the air at low exposure (Inoue et al., 2008). S-BMA can only be formed after inhalation of toluene, and then ethyl acetate would be rapidly hydrolyzed and mineralized by esterase. Hence, when the exposure is low, there would be no metabolism between organic solvents (Browning, 1965). These results suggested that as kitchen air toluene concentrations increase, so do urinary metabolite concentrations, and the weaker effect (r < 0.3) may be due to lower air toluene exposure concentrations (the mean value in this study's kitchen was below the standard limit). Another study showed a closer correlation between 2-OHN and ∑OHN in rural inhabitants, with a correlation coefficient of 0.978 (p < 0.01), indicating that even in the absence of 1-OHN data, 2-OHN was still a good indicator of the overall urinary naphthalene exposure levels (Fan et al., 2009). Thus, the internal and external exposure levels of benzene, toluene and naphthalene were all well correlated in this study's chefs and kitchens.

Table 4
Correlation analysis of kitchen's aromatic hydrocarbons and its urinary metabolites (n = 72).[a]

Correlation coefficient	Benzene (μg m^{-3})	S-PMA (μmol mol^{-1} Cr)
Benzene (μg m^{-3})	1.000	0.345**
S-PMA (μmol mol^{-1} Cr)		1.000
	Toluene (μg m^{-3})	S-BMA (μmol mol^{-1} Cr)
Toluene (μg m^{-3})	1.000	0.262*
S-BMA (μmol mol^{-1} Cr)		1.000
	Naphthalene (μg m^{-3})	2-OHN (μmol mol^{-1} Cr)
Naphthalene (μg m^{-3})	1.000	0.318**
2-OHN (μmol mol^{-1} Cr)		1.000

[a] *** p < 0.001, ** p < 0.01, * p < 0.05.

3.4. Health effect by benzene, toluene and naphthalene exposure

The concentration of kitchen pollutants and their corresponding urine metabolites showed a significant positive correlation, so their health effects on chefs were explored. FEV1/FVC, SBP, DBP, MDA, and 8-OHdG were monitored to evaluate lung function, blood pressure and oxidative stress levels. The results showed that the average level of FEV1/FVC was 0.73 ± 0.052 > 0.7, indicating that the chefs had no chronic pulmonary obstruction. The mean SBP and DBP values of chefs were obtained as 122.17 ± 14.70 and 77.58 ± 11.04 mmHg, respectively, which fall within the normal blood pressure range. Table 5 shows that benzene, toluene and naphthalene in kitchen cooking pollution have no significant effect on blood pressure. That is, benzene, toluene and naphthalene produced in kitchen may not damage the pressure and elasticity of the blood vessels or heart function of the chef.

MDA and 8-OHdG concentrations in urine and saliva have been widely applied as indicators of systemic oxidative stress (Cui et al., 2018). The mean concentrations of MDA in the chefs were obtained as 1968 ± 1237 μmol mol^{-1} creatinine in oral cells, 1868 ± 1167 μmol mol^{-1} creatinine in urine, and 1741 ± 1145 μmol mol^{-1} creatinine in saliva. The mean concentrations of 8-OHdG were 0.048 ± 0.031, 0.038 ± 0.021 and 0.032 ± 0.021 μmol mol^{-1} creatinine in oral cells, urine and saliva from chefs, respectively. Another study with chefs showed that urinary MDA concentrations ranged from 347.2 to 386.3 μmol mol^{-1} Cr and 8-OHdG concentrations ranged from 2.3 to 4.5 μmol mol^{-1} Cr, which were positively correlated with urinary levels of 1-OHP (Ke et al., 2016). In contrast, the results showed that the chefs in this study were severely affected by lipid peroxidation, while the degree of DNA damage was slightly lower, perhaps due to locality and cooking habits.

The effects of benzene, toluene and naphthalene in kitchen cooking pollution on oxidative stress levels are also shown in Table 5.

Table 5
Health effect of benzene, toluene, and naphthalene in chefs.[a,b]

	S-PMA		S-BMA		2-OHN	
Health indicators	Before	After	Before	After	Before	After
lg SBP						
lg DBP						
FEV1/FVC						
U-8OHdG					+*	+*
S-8OHdG			−*	−**	+*	+*
C-8OHdG			−*	−**	+*	+*
lg U-MDA						
lg S-MDA		+*		−*		
lg C-MDA						

[a] U-, S-, C- represent indicators in urine, saliva and oral epithelial cells, respectively.
[b] *** p < 0.001, ** p < 0.01, * p < 0.05; + positive effect; - negative effect. Blank value means no significant.

Naphthalene had only a significant positive correlation with 8-OHdG ($p < 0.5$), suggesting that the effect of naphthalene on the oxidative stress of these chefs may be caused by DNA damage rather than lipid peroxidation. Few studies have explored the relationship between exposure levels of PAHs and potential oxidative stress biomarkers of occupational exposed chefs. As reported by Ke et al., a positive relationship was also observed between urinary concentrations of 1-hydroxypyrene (1-OHP) and 8-OHdG, suggesting that exposure to pyrene or possibly other PAHs in cooking-oil fumes may cause DNA damage (Ke et al., 2009, 2016). However, a large amount of hazardous chemicals may be released during cooking, and co-exposure of these pollutants may produce a synergistic or antagonistic effect. For instance, the toluene metabolite S-BMA was significantly negatively correlated with 8-OHdG and may have similar antagonism with other benzenes. Kim et al. also found that the mixture of toluene and xylene had an antagonizing effect on oxidative stress (Kim et al., 2011). Consequently, the results of this study may be affected by the combination of benzene series or other substances, and systematic studies are further needed to elucidate the relationship between them as well as their potential health effects.

The effects of benzene and toluene on oxidative stress level became stronger after including kitchen substance concentration corresponding to urinary metabolites, while the effect of naphthalene was the same as before. In a study on the $PM_{2.5}$ component exposure and heart rate variability of taxi drivers before and after the Beijing Olympic Games, some effects were estimated to become more powerful after including the model $PM_{2.5}$ and components (Wu et al., 2011). In addition, there was a good correlation between the internal and external exposure levels of the above-mentioned substances. Hence, these results reinforce the idea that kitchen substance concentrations can affect oxidative stress levels by influencing exposure levels within the chefs.

3.5. Health implications

Steaming, frying and grilling are the most popular cooking methods in the world, which have different effects on indoor pollution and public health. This paper found that the relationship between internal and external exposure, kitchen exposure through the impact of internal exposure of chefs, and then cause health effects. The monitoring of aromatic hydrocarbons in commercial kitchens should be strengthened, especially the concentrations of naphthalene and benzene. It is recommended that commercial kitchens should establish an excellent ventilation system; introduce indoor air pollutant standards for kitchen environment, and at the same time, strengthen scientific and technological innovation in this field, and improve the mechanization of cooking operations.

Nevertheless, all volatile organic compounds were not considered in this study due to constraints (time, budget, subject availability, etc.), and only the positive or negative health effects due to kitchen exposure were discussed herein. The influencing factors of kitchen pollutant levels were not completely consistent, which may lead to some differences. However, this is the first complete systematic study to subdivide chef categories in order to evaluate external and internal exposure levels and their correlation, health effects including cardiopulmonary function and oxidative stress level, and the primary assessment of their health risk. These results provide a multi-angle reference for health assessment of the working environment of chefs. Future studies are warranted to explore efficient detection methods to assist the research on individual exposure of chefs, and to make calculations and evaluations more accurate based on larger samples and 24-hour follow-up research on chefs.

4. Conclusions

The findings of this work highlight the different effects of cooking styles on indoor air pollution in kitchens and the correlation between internal and external exposure. The concentration of naphthalene in the air of different kitchens is the highest, and the concentration of benzene is the lowest. The air concentration of toluene in the frying kitchen and benzene in the grilling kitchen exceeded the standard. Regarding the metabolites of pollutants in urine, the S-BMA of frying chefs is significantly higher than that of chefs of other cooking styles, which is consistent with the relatively high air concentration of toluene. And the internal and external exposure of pollutants has a good correlation, so the oxidative stress level is affected by 2-OHN and S-BMA, indicating the potential health risks of these occupational exposed chefs. For the three cooking styles: steaming, frying and grilling, more attention should be paid to the health risks of frying and grilling chefs. This study implies the necessity of monitoring typical aromatic hydrocarbons, as well as assessing their potential health effects in large-scale chef groups, and improving the ventilation in kitchens.

CRediT authorship contribution statement

Lei Huang: Methodology, Formal analysis, Writing - original draft. **Haonan Cheng:** Methodology, Formal analysis. **Shengtao Ma:** Methodology, Data curation. **Ruoying He:** Visualization, Investigation. **Jicheng Gong:** Visualization, Investigation. **Guiying Li:** Writing - review & editing. **Taicheng An:** Conceptualization, Supervision.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary material

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