

RESEARCH ARTICLE

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Comparing Differences in Blood Malondialdehyde and Risk Factors Among Four Occupations Around an Industrial Estate, Rayong Province of Thailand

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Abstract

Background: The chemical concentrations in the atmosphere from air quality monitoring stations of Map Ta Phut Industrial Estate, Thailand. **Objective:** This study aimed to compare blood levels of malondialdehyde (MDA) according to risk factors among participants in four occupations in the MTP-IE, Rayong Province. **Methods:** Interviews were conducted; urine samples were collected to analyze S phenyl mercapturic acid, 1,2 dihydroxy-4-(N acetyl) butane, and 1,2 dichloroethane twice on weekdays and weekends; blood samples were collected to analyze MDA in a sample of four occupational groups: traffic police, outdoor food vendors, public transport drivers, and fishery workers. **Results:** The study found that factors causing significantly different levels of MDA ($p < 0.05$) were 1,2 dichloroethane (mg/L) (weekend) ($p < 0.006$), BMI (kg/m²) ($p < 0.028$), occupation ($p < 0.001$), personal hygiene care (per week) ($p < 0.001$), and at-work physical activity ($p < 0.021$). **Conclusions:** To reduce blood MDA levels, it is recommended to monitor the health of workers in high-risk areas by examining MDA levels in the blood to assess the risk of cancer from exposure to 1,2 dichloroethane and to encourage workers to address personal hygiene.

Keywords: Carcinogen- Industrial Estate- Malondialdehyde- Rayong Province

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Introduction

Air pollution is now considered one of the greatest environmental risks to health. In 2019, the World Health Organization (WHO) found that 99% of the world's population lived in areas that did not meet air quality guidelines. The combined effects of air pollution were found to be associated with 6.7 million premature deaths per year. Ambient (outdoor) air pollution was estimated to be responsible for 4.2 million premature deaths worldwide in 2019 [1]. Therefore, attention should be paid to health in groups working outdoors in areas with air pollution. This is especially true of Thailand's Rayong Province, which is one of the provinces in the Eastern Economic Corridor (EEC) that has had air pollution problems. It was found that benzene, 1,3-butadiene, and 1,2 dichloroethane exceeded the standard values [2].

The statistics of cancer incidence are high. In 2022, the National Cancer Institute's Thailand Cancer Registry revealed that each year, Thailand has more than 140,000 new cancer patients, or approximately 400 people per

day [3]. The causes of cancer stem from many factors, including those within the body, such as genetics, and those outside the body, such as behavior, lifestyle, and environment, including smoking, drinking alcohol, eating food contaminated with carcinogens, and being exposed to carcinogens from one's occupation and the environment [4], such as industrial plants, chemical spills, transportation, agriculture, and household activities [5]. Ecological study has shown that increased levels of air pollution are associated with increased incidence of overall oral cavity and pharyngeal cancers in the US [4].

The health of people at risk can be supported by preventing exposure to carcinogenic substances such as benzene, 1,3-butadiene, and 1,2 dichloroethane via the environmental monitoring of the Pollution Control Department, which monitors chemical concentrations in the atmosphere from air quality monitoring stations. This research operation is in line with the spirit of the Occupational and Environmental Disease Control Act B.E. 2019, which established a mechanism for the surveillance, prevention, and control of occupational and environmental

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diseases for people who are or may be exposed to pollution [6], including at-risk occupational groups such as traffic police, outdoor food vendors, public transport drivers, and fishing workers. However, there is still a lack of sufficient information to protect this group's right to health care.

It is important to monitor the health of those with exposure to air pollution. Therefore, biomarkers of exposure [7, 8] and biomarkers of effect are important in the context of occupational and environmental health. This is especially true for healthcare professionals who may be exposed to various hazards. Biomarker assessment of oxidative stress from serum malondialdehyde (MDA) levels is widely used [9] for this purpose because there is a relationship between the level of exposure to carcinogenic substances such as benzene and MDA level [10].

MDA is a compound resulting from lipid peroxidation, a process caused by free radicals reacting with polyunsaturated fatty acids in cell membranes. MDA is considered an indicator of oxidative stress damage [11]. Many chemicals can cause oxidative stress, including the chemicals examined in this study, namely benzene, 1,3-butadiene, and 1,2 dichloroethane. A study of 12 urinary volatile organic compounds metabolites (mVOCs) and oxidative damage biomarkers (ODBs) in workers and children in e-waste recycling areas found that e-waste workers exhibited significantly higher levels of mVOCs exposure and ODBs than e-waste children and control adults [12]. When MDA is generated in large quantities, it can react with proteins and DNA, causing damage to cells and potentially leading to various diseases [11], such as kidney disease, brain disease [13], and cancer [14]. A study found that gas station workers exposed to benzene experienced oxidative stress, leading to the expression of proteins linked to cancer pathways [15].

Currently, there is a lack of in-depth supporting data indicating the carcinogenicity of some potential carcinogens. The researchers therefore see a research gap regarding knowledge of the exposure to carcinogenic substances, namely benzene, 1,3 butadiene, and 1,2 dichloroethane in urine, and their impact on MDA. Therefore, the researchers aimed to monitor health and compare differences in blood MDA and risk factors among four occupations around an industrial estate in Rayong Province. The results of the study yielded information that provided guidelines for screening the health of outdoor workers to ensure equal coverage of those at-risk workers.

Materials and Methods

Study Sample

In this quantitative study, data were collected from the analysis of samples obtained from four occupational groups, namely, traffic policemen, outdoor food vendors, public transport drivers, and fishery workers who worked around the Map Ta Phut Industrial Estate. This study employed a calculation formula using G*Power (version 3.1.9.7) to set the effect size to 0.15, the α error value to 0.05, and the power $1-\beta$ value to 0.84 as well as establish the primary variables by making predictions according to the conceptual framework. The sample size was 50 people from each of four occupational groups—traffic police,

outdoor food vendors, public transport drivers, and fishery workers working in the Map Ta Phut Industrial Estate for a total of 200 people.

This study employed the following inclusion criteria: (1) individuals working in one of the four occupations outdoor food sales, traffic policemen, public transport drivers, and fishery workers—as their main job in the Map Ta Phut area for 3 months; (2) those aged 18–60 years; and (3) those who voluntarily cooperated as research volunteers. The exclusion criteria included individuals who could not participate as research volunteers until the research project was completed. In addition, individuals with serious illnesses such as kidney and liver disorders and severe diabetes were excluded. This study was approved by the Human Ethics Committee of Burapha University (number HS 096/2023) before data collection. Voluntary expression of consent by a study subject and sufficient information disclosure about the research are essential elements of the informed consent process.

Research Tools

Four types of research tools were used in this study: (1) the interview form, which consisted of eight items of demographic, social, and health behavior information, including gender, age, smoking, drinking alcohol, body mass index (BMI), occupation, overtime work, and work characteristics; (2) equipment for collecting urine samples, including plastic cups and a 50 mL polyethylene jar; (3) blood collection equipment, including alcohol, cotton balls, gloves, tourniquets, blood collection sets with sizes 21 and 22 needles and blood sampling tubes coated with EDTA, anti-coagulant and heparin, size 5 or 10 cc for oxidative stress and MDA analysis.

Data Collection

1. Subjects were interviewed by the research team. The interviews were conducted at appointed locations for each occupation that were conveniently located, such as the community hall, public bus resting points, police stations, etc.

2. The urine samples were collected on two occasions during a workday and a weekend. It was recommended that the participants collect the samples at the end of a work shift by collecting mid-stream urine in a plastic cup, then pouring at least 50 mg or half a jar of urine into a polyethylene jar. When the researchers obtained a urine sample, they quickly packed the sample in a box with a temperature below 4°C and transported it to the laboratory to analyze S-phenyl mercapturic acid for benzene (S-PMA), 1,2 dihydroxy-4-(N acetyl) butane for 1,3 butadiene (1,2-DDB) and 1,2 dichloroethane in urine (1,2-DCE).

The analytical process was performed to evaluate 1) S-PMA: The instrument standards based on the vendor's standard according to Angerer et al. method [16], 2) 1,2-DDB: the analysis was performed according to the standard of the instrument according to Anttinen-Klemetti et al. [17], and 3) 1,2-DCE: the researchers collected urine samples to analyze dichloroethane in the laboratory. The analysis process was performed according to the steps of Payan et al. [18]. The standard of the instrument was

based on the standard calibration methods of the vendor company.

3. Subjects' morning blood samples were collected at the same appointments as the urine collection. Those responsible for drawing blood included medical technicians and nurses. Blood was drawn from a vein in the inner crook of the arm using a vacuum tube with a volume of 8–10 mL. The researcher then packed the sample in a box at temperatures below 4°C and sent it to the laboratory for analysis according to the steps of the instruction manual for the HPLC analysis of MDA in plasma/serum (order number 67000, Germany) [19]. Internal quality was controlled using two levels of plasma. The instruments were calibrated following the vendors' standard methods. The lowest limit value of MDA in plasma was set as lower than the LOQ, which is 0.72 µg/L. The reference value was < 36 µg/L.

Data Analysis

Analysis of the data distribution using Kolmogorov–Smirnov statistics showed that the data were non-normally distributed. The analysis included (1) descriptive analysis: the median range and percentile rank were used to analyze urinary metabolites, including S-PMA, 1,2-DDB, 1,2-DCE, and MDA in blood, and (2) inferential statistics tests, consisting of (a) MDA level comparisons classified according to quantitative variables such as age (years), body mass index (BMI) (kg/m²), income (baht per month), number of cigarettes per day, period of smoking (years), number of smoking days per week, amount of liquor consumed (glasses), S-PMA, 1,2-DDB, and 1,2-DCE with Kruskal–Wallis test statistics and (b) comparison of gender, smoking, period of service, personal hygiene, and performance characteristics using the Chi square test with statistical significance set at $p < 0.05$.

Results

Indicator of Oxidative Stress: Malondialdehyde (MDA)

The study results revealed that the concentration levels of Malondialdehyde (MDA) in the blood of the sample group, which consisted of four occupational groups: traffic police, street food vendors, public transport drivers, and fishermen around the Map Ta Phut Industrial Estate, mostly fell within the 75th percentile, with 75 individuals (37.5%) in that range, as shown in Table 1.

Comparison of MDA in the blood among members of four occupational groups

The results of the comparative analysis of percentile rank of MDA in the blood of overall samples in four occupational groups, including traffic police, outdoor food vendors, drivers for hire, and fishing workers who work outdoors around the Map Ta Phut Industrial Estate, were classified according to general information, smoking history, history of drinking, and metabolites in urine. The study found that the following factors led to statistically significant differences in the level of MDA ($p < 0.05$): BMI (kg/m²) ($p < 0.028$), income (baht/month) ($p < 0.002$), and 1,2 dichloroethane in urine (weekend) ($p < 0.006$), as shown in Table 2.

The percentile rank of S-PMA, 1,2-DDB, and 1,2-DCE was compared by quartiles of increasing levels of MDA in the blood among 200 subjects. The study also found that occupation ($p < 0.001$), personal hygiene (days per week) ($p < 0.001$), occupation ($p < 0.009$), and exertion at work ($p < 0.021$) significantly affected blood MDA levels, as shown in Table 3.

Discussion

Comparison of malonaldehyde (MDA) levels in the blood classified by demographic factors (gender and age)

This study did not find differences in MDA levels by gender. MDA is a marker of antioxidant activity in oxidative stress in cancer patients [20]. The concentration of MDA in plasma did not exceed the reference value of 36 µg/dL analyzed by HPLC (Chromsystem), which is inconsistent with the results of Nielsen et al. [21], who found that the concentration of MDA in the blood increased according to frailty ($p < 0.001$), which reflects further differences between men and women, namely, females lose the stimulating effect of estrogen on antioxidant enzymes after menopause [22]. However, this study did not find that MDA increased in females, probably because the majority of the sample was male. Therefore, no change in MDA was seen [23]. Although the study did not find that gender has a significant effect on increasing MDA levels, health should be continuously monitored in both males and females in at-risk groups.

This study's results did not show differences in MDA levels by age, probably because the sample had a mean (standard deviation) age of 47.44 (13.861) years.

Table 1. Number and Percentage of MDA Concentration Levels in Blood

MDA	Traffic police		Outdoor food vendor		Public transport driver		Fishing worker		Overall	
	(n = 50)		(n = 50)		(n = 50)		(n = 50)		(n = 200)	
	n	%	n	%	n	%	n	%	n	%
MDA (nmol/L)										
Percentile 25	10	20	10	20	18	36	26	52	64	32
Percentile 50	17	34	19	38	16	32	9	18	61	30.5
Percentile 75	23	46	21	42	16	32	15	30	75	37.5
Mean±SD	97.58±28.11		88.34±15.68		89.98±30.00		80.34±21.63		89.09±25.09	
GM±GSD	1.97±.11		1.94±.07		1.93±.11		1.89±.10		1.93±.10	
Median (Min-max)	83 (56-167)		83 (56-139)		83 (69-236)		69 (56-180)		83 (56-236)	

Abbreviations: MDA, (Malondialdehyde); SD, (Standard Deviation); GM, (Geometric Mean); GSD, (Geometric Standard Deviation).

Table 2. Percentile Rank of SPMA, 1,2-DDB and 1,2-DCE in Urine by Quartiles of Increasing Levels of and MDA in the Blood

Factors	MDA (nmol/L) (n=200)								p
	PR 1†		PR 2‡		PR 3§		PR 4		
	ME	PR	ME	PR	ME	PR	ME	PR	
Demographic									
Age (Years)	51	25	48	22	47	19	50	20	0.294
BMI (kg/m²)	23.3	5.4	24.7	6	24.6	6.5	25.1	5	.028*
Income (USD per month)	439.1	292.7	585.4	395.1	585.4	439.1	585.4	439.1	.002*
Cigarettes (number per day)	10	5	8	15	10	6	10	12	0.963
Alcohol (glasses per day)	3	3	4	3	3	3	3	3	0.967
Urine of weekday									
S-PMA (µg/g Cr)	0.0084	0.0001	0.00837	0.0001	0.0084	0.0001	0.0084	0.0001	0.696
1,2-DDB (mg/L)	0.091	0.116	0.08	0.105	0.046	0.088	0.091	0.066	0.165
1,2-DCE (mg/L)	0.465	0.167	0.488	0.18	0.505	0.15	0.471	0.152	0.454
Urine of weekend									
S-PMA (µg/g Cr)	0.0084	0.0001	0.0084	0.0001	0.0084	0.0001	0.0084	0.0001	0.578
1,2-DDB (mg/L)	0.423	0.145	0.462	0.132	0.471	0.146	0.452	0.121	0.357
1,2-DCE (mg/L)	0.207	0.205	0.138	0.154	0.114	0.305	0.126	0.113	.006*

Abbreviations: ME (Median); PR (Percentile Rank); MDA (Malondialdehyde); BMI (Body Mass Index); S-PMA (S-phenylmercapturic acid); 1,2-DDB (1,2 Dihydroxy-4 (N acetyl) Butane); 1,2-DCE (1, 2 dichloroethane). † Median of MDA of percentile rank (PR1) was 69; ‡ Median of MDA of percentile rank (PR2) was 83; § Median of MDA of percentile rank (PR3) was 97; || Median of MDA of percentile rank (PR4) was 111; *Statistically significant at p<0.05, analyzed using Kruskal Wallis test

Table 3. Percentile Rank Comparison of MDA Levels in the Blood of Overall Samples in Four Occupational Groups Surrounding the MTP-IE Classified According to Various Factors

Factors	MDA (nmol/L) (n=200)								p
	PR 1		PR 2		PR 3		PR 4		
	n	%	n	%	n	%	n	%	
Sex									
Male	43	67.2	41	67.2	22	56.4	26	72.2	0.515
Female	21	32.8	20	32.8	17	43.6	10	27.8	
Cigarette smoking									
No	47	73.4	54	88.5	29	74.4	27	75	0.154
Yes	17	26.6	7	11.5	10	25.6	9	25	
Working duration (years)									
< 22	48	75	42	68.9	34	87.2	28	77.8	0.214
≥ 22	16	25	19	31.1	5	12.8	8	22.2	
Occupations									
Traffic police	10	15.6	17	27.9	7	17.9	16	44.4	<.001*
Outdoor food vendor	10	15.6	19	31.1	15	38.5	6	16.7	
Public transport drivers	18	28.1	16	26.2	6	15.4	10	27.8	
Fishing worker	26	40.6	9	14.8	11	28.2	4	11.1	
Personal hygiene (Days per week)									
0-4	27	42.2	12	19.7	7	17.9	8	22.2	.011*
5-7	37	57.8	49	80.3	32	82.1	28	77.8	
Standing to work									
Yes	16	25	31	50.8	21	53.8	13	36.1	.007*
No	48	75	30	49.2	18	46.2	23	63.9	
Exertion in work									
Yes	25	39.1	21	34.4	18	46.2	5	13.9	.021*
No	39	60.9	40	65.6	21	53.8	31	86.1	

*Statistically significant at p <.05, analyzed using Chi-square test

Normally, steady-state MDA peaks between the ages of 50 and 55. The variation in plasma MDA in older people is 12% [22]. Adults (aged 25–65 years) had lower plasma MDA concentrations than elderly people (aged ≥ 65 years) [13], but most of this study's subjects were not elderly. Although the study did not find significant differences in MDA according to age, it is still recommended to monitor continuously the health of MDA levels in the elderly at-risk group.

Body mass index

Based on this study's results, we recommend a campaign to encourage at-risk groups to reduce their BMI to normal levels by engaging in exercise [24] and controlling diet, focusing on fruits such as apples and vitamin C [25] because apples play a role in preventing superoxide radicals by inhibiting xanthine oxidase by increasing the heme oxygenase-1, an antioxidant enzyme in the immune system [26], to reduce MDA concentrations and the occurrence of oxidative stress and inflammation, which it is believed will help prevent cancer in the future.

Smoking and drinking

This study found that most of the sample did not smoke but did drink alcoholic beverages. Therefore, changes in MDA levels may not yet be seen. Drinking alcoholic beverages along with smoking can form adducts with many types of proteins and DNA. In the past, it was found that MDA levels were increased among groups that engaged in drinking alcohol and smoking [27]. The combined effect of smoking and drinking alcohol can lead to increased levels of damage to cell function [28]. This study did not analyze the combined effect of smoking and drinking on MDA; although the results were not significant, public health personnel in the area should campaign for those in at-risk occupations to reduce or stop smoking and drinking alcohol in order to prevent the risks associated with oxidative stress levels in the long term.

Chemicals

It was found that this substance increased MDA levels in the blood, indicating a risk of oxidative stress, while 1,3 butadiene had no significant effect on MDA in the blood. This study's findings suggest that MDA should be used as an indicator of oxidative stress from exposure to 1,2-dichloroethane [29, 30] but cannot be used as an indicator of benzene and 1,3-butadiene exposure. For those working in the four studied occupations, there is a chance of being exposed to this substance, mainly through the respiratory tract [30]. The level of severity increases with the amount of the chemical [29]. Therefore, local medical professionals should advise all risk level groups to take care of personal hygiene and wear appropriate masks. This study found that benzene is not a risk factor for increased MDA levels, as mentioned above. The results are inconsistent with previous studies indicating that higher concentrations of benzene were associated with increased MDA after adjusting for age, gender, and smoking, and superoxide dismutase (SOD) was significantly increased (4.49 and 3.54 times, respectively) at medium levels of t,t-MA compared with low levels of

t,t-MA ($p = 0.01$ and 0.034 , respectively) [31]. However, this study classified MDA levels according to the percentile rank of S-PMA.

Occupation

This study found that police professionals had the highest MDA level of the occupations studied, followed by drivers for public transport drivers, outdoor food vendors, and fishery workers, who had the lowest MDA levels due to the nature of their work environment and related activities. The traffic police profession is risky in terms of MDA level [32] because of the possibility of exposure to exhaust fumes from vehicles during rush hour. Therefore, these workplace conditions can expose officers to carcinogenic substances emitted from gasoline and diesel exhaust. In addition, they are exposed to air pollutants from industrial plants. This increases MDA levels in the blood [33]. Results from previous studies indicate an increased risk of cancer in traffic police [32]. Therefore, it is necessary to take serious measures to prevent the increased danger of traffic police being exposed to carcinogens. Public transport drivers may be exposed to non-carcinogenic and carcinogenic substances from diesel exhaust from buses [34], which contains harmful substances such as benzene, formaldehyde, polycyclic aromatic hydrocarbons (PAHs), and particulate matter, as well as from traffic conditions and air pollution (similar in this way to traffic police) [35].

Outdoor food vendors may face exposure to carcinogens through high-temperature cooking processes, particularly those that involve open flames or grills. These cooking methods can produce carcinogenic compounds such as PAHs and heterocyclic amines (HCAs); moreover, working in areas with heavy traffic or industrial activities can increase exposure to air pollution [36]. Fishery workers had the lowest MDA levels in this study even though this occupation is exposed to many hazards, including carcinogens from the use of chemicals in cleaning and maintaining fishing equipment [37], pollution in water sources [38], etc. Occupations have different working characteristics, resulting in different exposure to the carcinogens benzene, 1,3 butadiene, and 1,2 dichloroethane. These substances may cause oxidative stress, increasing MDA levels [39]. Public health personnel should monitor the health of at-risk occupational groups that are likely to be chronically exposed to carcinogens, such as traffic police, outdoor food vendors, public transit drivers, and fishermen working around industrial estates.

Personal hygiene

This study found that the frequency (per week) of personal hygiene care made a difference in the levels of MDA in the blood that were statistically significant ($p < 0.05$), which is in reverse causality. However, medical personnel in the area of high-risk workers should be encouraged to take good care of their personal hygiene in terms of both the frequency of hand washing and the use of masks. In addition, programs should be organized to provide sustainable health care to at-risk groups [36]. More attention should be paid to setting policies and

launching campaigns to increase engagement in personal hygiene care for at-risk groups to prevent exposure to carcinogens and oxidative stress [40].

Physical activity

This study found that increased physical activity reduces MDA; we defined physical activity as “activity at work that requires exertion or standing” ($p < 0.001$), which was probably due to the effects of physical exertion. Regular exercise can control the antioxidant enzyme system, which may delay increase in oxidative stress [41]. Additionally, physical fitness levels may affect antioxidant activity and MDA levels if regular exercise is a means for resisting oxidative stress [42]. However, this study did not evaluate the subjects’ exercise frequency. Therefore, high-risk groups should be encouraged to get more physical activity to reduce the chance of oxidative stress.

One strength of this study is that MDA level assessment in the blood of high-risk groups living in polluted areas offers insights into oxidative stress and the health effects of carcinogen exposure by evaluating metabolite levels in the urine of samples taken from subjects working in densely populated urban areas surrounding industrial estates. The study area has high levels of emissions from vehicles and industrial factories, which can lead to oxidative stress in exposed individuals. In addition, this study addressed important confounding variables. It has been noted previously that foods containing sorbic acid (e.g., shrimp paste, sausage, stuffed bread, canned food, and fermented fruits) and smoking can affect chemical metabolism [15, 43, 44]. This study found that food and urine metabolism were not different. However, smoking is a form of direct exposure to chemicals, and the study requested that subjects provide detailed information regarding their smoking habits, including number of cigarettes, smoking frequency, and smoking duration, to analyze their association with MDA.

This study supports the recommendation of establishing a health policy. It presented evidence that MDA levels in the blood can be used to assess the risk of cancer from exposure to 1,2 dichloroethane. Additionally, we recommend a campaign to encourage high-risk groups to maintain their BMI at a normal level. Moreover, physical activity and strict personal hygiene should be advised to reduce the levels of MDA, thereby reducing oxidative stress.

This study also suggests that policymakers should implement strict pollution control measures to reduce the health risks associated with oxidative stress. In addition, a public healthcare program should be organized with public health strategies that focus on the needs of vulnerable groups of outdoor workers, namely traffic police, outdoor food vendors, public hire drivers, and fishermen working around the Map Ta Phut Industrial Estate, who live in highly polluted areas. However, workers may be exposed to multiple chemicals (multi-chemical) from various important sources such as industry, traffic, and everyday chemicals that may affect MDA in the future. Therefore, it is necessary to study the mutual associations between independent variables. This study is a preliminary approach that assesses the exposure to the

most contaminants in the environment and is of greatest concern because the chemicals studied can cause cancer.

In conclusions, the study found differences in the amounts of chemical metabolites present on weekdays and weekends. Furthermore, it was discovered that on weekdays, there were substantial differences in blood MDA levels based on 1,2 dichloroethane (mg/L) levels, BMI, occupation, personal hygiene, and levels of at-work physical activity. It should be advised that variables associated with risk should be regulated to prevent oxidative DNA damage and other inverse factors, such as physical activity; however, this study has some limitations.

Author Contribution Statement

All authors contributed equally in this study.

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Conflict of interest

The authors have no conflicts of interest associated with the material presented in this paper.

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