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Impacts of In-Cabin Exposure to Size-Fractionated Particulate Matters and Carbon Monoxide on Changes in Heart Rate Variability for Healthy Public Transit Commuters

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Abstract: To evaluate the cardiovascular impact of traffic-related pollutant exposure on healthy young adults, the research team has collected the primary data of in-cabin exposure to air pollutants and heart rate variability (HRV). Twenty young healthy college students were recruited in Taipei metropolitan area. In addition to electrocardiogram, personal exposure to air pollutants, i.e., particulate matter (PM) and carbon monoxide (CO), and weather conditions, including temperature and relative humidity (RH), on campus, bus, and mass rapid transit were monitored continuously. The following HRV parameters were evaluated using generalized additive mixed model to adjust for personal and meteorological variables: heart rate (HR), the square root of the mean of the sum of the squares of differences between adjacent normal-to-normal (NN) intervals (r-MSSD), the standard deviation of all NN intervals (SDNN), the percentage of successive NN interval differences greater than 50 ms (pNN50), low-frequency power (LF), high-frequency power (HF), total power (TP), and LF/HF. They were assessed to find out the association between in-cabin exposure and HRV parameters. Compared with the HRV parameters measured on campus, the percent changes in r-MSSD, SDNN, pNN50+1, LF, HF, and TP decreased when the participants were in public transits. After adjusting for all locations, 5 min moving averages of $PM_{2,5-10}$ and PM_1 were significantly associated with the increase in the percent changes in HR and SDNN. Additionally, 5 min moving averages of PM_{2.5-10} exposure were significantly associated with the decrease in the percent change in HF, while it was significantly associated with the increase of the percent change in LF/HF. The reduction of the percent change in HR was also found to be significantly associated with 5 min CO moving averages. To conclude, current analyses have shown that size-fractionated PMs and CO exposure in public transits might lead to significant changes of HRV parameters for healthy young adults.



Keywords: particulate matter; carbon monoxide; heart rate variability; public transit; in-cabin exposure

1. Introduction

In the past few decades, industry, transportation, and economy in Asian countries have developed and even flourished. However, the air quality of human settlements has deteriorated severely at the same time. According to the report of the World Health Organization (WHO), air pollutants exert a great impact on human beings. According to estimates, in 2010, there were about 3.2 million premature deaths caused by exposure to fine particulate matter (particulate matter with an aerodynamic diameter less than 2.5 μ m, or PM_{2.5}). The premature deaths were mainly caused by cardiovascular diseases (CVD), and more than half of these cases occurred in fast-growing Asian countries [1].

Air pollution exposure can be viewed as a function of the concentration of contaminants in a microenvironment and the time an individual is exposed to the microenvironment. In industrialized countries, particulate matter (PM) generated by traffic pollution accounts for a significant proportion of total particulate emissions [2]. Air pollutants in traffic-related microenvironments are undoubtedly mainly derived from traffic emissions. Therefore, commuters are often exposed to high concentrations of pollution that do not meet air quality standards. In fact, traffic-related air pollution exposure intensity varies with routes, traffic loads, and commuting modes [3–7]. Previous studies have indicated that commuters using engine vehicles, i.e., cars, trains, subways, trams, or buses, have higher air pollution exposure than those who adopt active commuting, i.e., walking or cycling [8]. Furthermore, previous studies have found that pedestrians and cyclists have higher PM exposure on high-traffic routes when compared with on low-traffic routes, while commuters travelling in diesel-powered buses have three to four times the PM exposure in comparison with gasoline-powered bus commuters [9–12].

Furthermore, many epidemiological studies have shown that particulate air pollutants can cause cardiovascular adverse reactions, increase mortality from cardiac failure caused by the exposure to PM, and face an increasing risk of ventricular arrhythmias, and that the changes in blood pressure are associated with the changes in heart rates (HR) [13–17]. Previous scientific studies have pointed out that autonomic nervous system (ANS) dysfunction may be an important pathway leading to cardiovascular diseases. The ANS contains the sympathetic nervous systems (SNS) and the parasympathetic nervous systems (PNS). When the SNS is stimulated, the heart rate will increase. Conversely, when the PNS is stimulated, the heart rate will decrease. If the SNS and PNS are unbalanced, the heart contraction will be affected and cardiovascular disease symptoms may appear [13,18]. The heartbeat of a human body has a considerable degree of variation even when it is in a stable state. In recent years, the analysis of heart rate variability (HRV) has become an important method for assessing cardiovascular functions and ANS regulation. The HRV measurement is relatively simple and feasible, so many environmental epidemiological studies adopt the HRV to explore the relationship between environmental air pollutants and human cardiovascular health. Too low HRV represents reduced dynamic complexity, adaptability, and ability to overcome continuous changes in the environment [16,19,20].

Panel studies have been adopted to investigate the short-term effects of outdoor PM pollution across a wide range of environmental settings [21]. Although previous studies have found that PM is associated with the changes in HRV, and the effect of particulates on HRV changes varies with particle sizes [20,22,23], current research into the effects of particulate exposure on cardiovascular functions and those of ANS on public transit is still limited, especially for healthy young populations [16,24]. Therefore, this panel study used HRV to assess the effects of traffic-related air pollution on cardiovascular functions, by simultaneously monitoring PM and carbon monoxide (CO) exposures as well as HRV on healthy young commuters who are exposed to size-fractionated PMs and CO on public transit.

2. Experiments

2.1. Study Design

The participants of this study were on-campus boarding students. Announcements were posted on campus to recruit volunteers who did not smoke, drink, and have no medical history of heart diseases, hypertension, and asthmatic diseases. In order to reduce the possible errors in the study, in addition to requiring the participants to follow normal routine activities, they were required to work and rest as usual, not to drink beverages containing caffeine or tea, and not to exercise vigorously or stay up late during the test period. After obtaining the consent of the volunteers, they were asked to sign the consent form before sampling. This study was approved by the ethics committee of Fu Jen Catholic University (approval number C9728).

Before the implementation of the monitoring, the researchers taught the participants to wear the electrocardiogram (ECG) record analyzer and personal pollution monitors, and asked them not to leave the campus on the first day, and to take a bus with the researcher the next day. Personal pollution monitors were worn in the breathing zone. All participants were instructed to keep the personal pollution monitors with them at all times. In this way, the researchers could compare the different impacts of PM and CO exposures on the HRV. In addition, the participants were required to record a personal time–activity log during the observation period; they were expected to record the activities on campus at intervals of 30 min, including the meal time and the start and end time of the place in which they arrived and they left. In the end, a total of 26 participants aged between 18 and 27 were recruited to take part in the study. Due to the loss of monitoring data caused by human error and meteorological factors during the monitoring period, the data for 20 out of 26 participants were statistically analyzed. Among them, ten were male while the remaining ten were female.

The study was conducted from 19 May 2009 to 4 March 2010. The monitoring period for each participant lasted two consecutive days: on the first day the relevant personal indicators were monitored on campus, and the next day participants took mass transit so that pollution exposure as well as HRV on public transportation could be monitored. As a result, the monitoring period started from 7 am on the first day and ended at 5 or 6 pm the next day. The bus and the Taipei metro/subway system (Mass Rapid Transit, MRT), the commonly seen public transportation, were used as the commuting vehicle where the study was sampled. The researcher met with the participants at 8 am the next morning, and they took the bus and MRT together. The selected bus route started from Fu Jen University Station, passed Zhongzheng Road to Dahan Bridge, and ended at Xinpu Station. The whole route was about five kilometers long, and it took about 20 to 30 min by bus. Next, the participants and the researcher took the MRT Bannan Line from Xinpu Station to the Taipei City Hall Station, which took about 20 min. After arriving at the Taipei City Hall Station, they rested in the station for half an hour and took the MRT to Xinpu Station, and then returned to the campus on the original bus route (Figure 1). During the commute, the researcher also recorded the time-activity logs, including the start and end of walking and riding, the environmental conditions, and the physical discomfort, if any, of the participants. The total monitoring time of each case was 30 ± 2 h. If the weather conditions were unstable on the way, such as raining, the sampling was canceled and the monitoring time was rearranged.



Figure 1. Maps of the different transit routes across Taipei Metropolitan Area. Image taken from Google Maps.

2.2. HRV Indices

In this study, the HRV parameters were used as cardiovascular function and ANS evaluation indicators. The portable ECG recorder and analyzer (model E3-8010, MSI, Taiwan) was adopted for monitoring. The sampling rate was 250 Hz. There are two approaches to measure HRV: analysis in the time or in the frequency domain. These measures are based on the analysis of interbeat intervals of normal beats determined from an electrocardiogram. Time domain analysis addresses the question of variability, while frequency domain analysis addresses the question of the underlying rhythms [25]. The time domain parameters of HRV included the square root of the mean of the sum of the squares of differences between adjacent normal-to-normal (NN) intervals (r-MSSD), standard deviation of all normal-to-normal intervals (SDNN), and NN50 count divided by the total number of All NN intervals (pNN50). The frequency domain HRV parameters included power in the low-frequency range (LF), power in the high-frequency range (HF), total power (TP), and LF/HF. Among the HRV parameters, SDNN is a time domain measure of overall HRV; HF and LF are representative of predominantly parasympathetic and sympathetic autonomic cardiac regulation. The LF/HF ratio represents the relative balance between sympathetic and vagal nervous activities [25]. Together with HR, there were eight HRV parameters in total. Data of these eight parameters were averaged for each five minute monitoring segment.

2.3. Air Pollution and Weather Conditions

Exposure to PM of different particle sizes and the changes in environmental temperature and relative humidity (RH) were measured with a DUST-check portable dust monitor (series 1.108, Grimm, Germany). The equipment uses light-scattering effects to distinguish the particle size and quantity, and the count value is converted into mass concentration through the calculation of dust mass distribution. A total of 15 particle sizes can be measured, which range from 0.3 μ m to 20 μ m. The detection limit for mass concentration is 0.1 μ g/m³, and the sampling rate reaches 1.2 L/min. The monitor's meteorological sensor can simultaneously obtain temperature and humidity data. In this study, the mass concentrations of PM₁₀ (particulate matter with an aerodynamic diameter less than 10 μ m), PM_{2.5}, and PM₁ (particulate matter with an aerodynamic diameter less than 10 μ m) were selected, and the data were set to be output per minute. Mass concentrations of PM_{2.5-10} (coarse particulate matter) were obtained by subtracting the PM_{2.5} fraction from the concurrent PM₁₀ levels. A similar approach was applied to derive the data for PM_{1-2.5} concentrations, an intermediate mode between coarse and fine particulate matter. In addition, we used an electrochemical gas sensor (Dräger PAC III CO detection

instrument, Dräger, Germany) to detect CO concentrations, where the equipment has a detectable range from 0 ppm to 2000 ppm, with the detection limit of 1 ppm. The data were set to output one value for CO concentrations per minute as well. In addition to the routine calibration and maintenance of the instrument used in this study, the researcher performed essential calibration of instrument readings and flows before and after monitoring.

2.4. Statistical Analysis

After the participants' pollutant exposure data and HRV data were documented in Excel, complete data from the two-day monitoring were included in the analysis, and descriptive statistics were performed with SPSS 12.0. Based on the sampling logs, the data were classified with three major locations: bus, MRT, and campus environment. In order to increase the normality and stability of the HRV parameters, natural logarithm transformation (ln) was performed to HR, r-MSSD, SDNN, pNN50, LF, HF, TP, and LF/HF. In addition, since a part of pNN50 values were 0, one point had been added to all pNN50 values before logarithmic transformation was performed, for the addition of 1 could help the natural logarithmic transformation of pNN50 to perform smoothly [16]. Using the Shapiro–Wilk test, the participants' air pollution exposures, ln-transformed HRV, and meteorological conditions were found to be normally distributed. Since the exposure data at different locations showed non-normal distribution, the Mann–Whitney test was used to compare the in-cabin and on-campus exposures. In the study, statistical significance was inferred at a *p* value of 0.05.

At present, most environmental epidemiological studies often use a linear model to analyze the impact of air pollution on human health [22,26–30]. Since the participants of this research were all independent individuals, the missing data could not be processed. Moreover, measurements were made repeatedly over time, and the interference factors of the monitoring time, temperature, and humidity are in fact nonlinear to the HRV. Consequently, it was not appropriate to analyze the results with a linear model. On the other hand, a mixed model can handle the problem brought by repeated measurement and treat the missing data as a random loss, so the aforesaid problems will not be analysis difficulties for a mixed model. If a linear mixed model (LMM) is applied, repeated measurement and missing data can be dealt with, but it is difficult to handle the problem of high nonlinearity. Therefore, this study used the generalized additive mixed model (GAMM) to solve the integration and analysis of repeatedly measured data, including the nonlinear effects, the treatment of missing data, and the individually different susceptibilities [31–33]. The overall model is given in the form:

$$y_i = \mathbf{X}_i \boldsymbol{\beta} + f_1(\mathbf{x}_{1i}) + f_2(\mathbf{x}_{2i}) + \dots + \mathbf{Z}_i \mathbf{b} + \varepsilon_i, \tag{1}$$

where y_i is the HRV-related parameter, X_i is a fixed-effects design matrix, β is a fixed-effects vector, f is the smooth function of the covariate x_k , Z_i is a random-effects design matrix, **b** is a random-effects vector, and ε is the residual covariance matrix.

In the current study, three time periods using moving averages, namely 5, 30, and 60 min, were adopted to evaluate the effect of the moving average of size-fractionated PMs and CO concentrations on participants' HRV. After monitoring data were removed if the activity log showed the personal monitors were not with the participant, the R statistical software 2.10.1 was applied, and the GAMM was used to model exposures to PM and CO in the three time periods with the heartbeat and HRV indices [31–33]. The independent variable is the average concentrations of particulate matter and carbon monoxide for various moving average times. The dependent variables are HRV-related parameters, including ln r-MSSD, ln SDNN, ln pNN50+1, ln LF, ln HF, ln TP, ln LF/HF, and ln HR. The time scale of this study is that when pollutant data were analyzed, the average value of pollutants at different timings was tracked backward in time by each pollutant exposure at the same time point as HRV. In this study, the data from the exposure to actual environments was used to observe the human body's reaction when humans were affected by $PM_{2.5-10}$, $PM_{1-2.5}$, PM_1 , and CO exposures [26]. With regard to random effect of the model, the study controlled the parameter of the participants, and by referring to

previous researches, listed gender, body mass index (BMI), activity locations (campus/bus/MRT/sleep), temperature, RH, time, and day of week as parameters of fixed effects. Such mixed-effects models have the advantage of adjusting for invariant variables by fixed-effects models and accounting for individual difference by random-effects models. The final models adjusted for smooth function terms in generalized additive models as fit by penalized cubic regression spline to reflect possible nonlinear effects of continuous covariates, including time, temperature, and relative humidity. In the analysis, data collected from the campus served as the benchmark group, and the coefficient representing the activity location in the model is a relative variation in comparison with the data from the campus. In order to observe the influence of pollutants on a certain physiological index, the results were expressed with a pollutant coefficient (β) that had been obtained through model analysis using the formula: ($e^{\beta} - 1$) × 100 [16].

3. Results

3.1. Summary Statistics of the Participants

Among the 20 participants, ten of them were male students while the remaining ten were female ones. The subjects were aged 21.8 ± 2.4 years (arithmetic mean \pm standard deviation (SD)), with BMI of 21.7 ± 3.6 kg/m². The round trip by bus took 74.0 ± 15.6 min, while the round trip by MRT took 86.1 ± 14.1 min. All the participants in the study did not smoke and did not have a history of heart diseases, hypertension, and asthmatic diseases. Since they were on-campus boarding students, they did not use household kitchen ranges. Also, based on the activity log records, they were not exposed to environmental tobacco smoke.

The data of the 20 participants' exposures to PM and CO were further compiled, and the average values were collated every five minutes (Table A1). The exposure concentrations of PM₁₀, PM_{2.5–10}, PM_{2.5}, PM_{1–2.5}, PM₁, and CO were $45.4 \pm 50.6 \ \mu\text{g/m}^3$, $16.3 \pm 37.1 \ \mu\text{g/m}^3$, $29.0 \pm 23.8 \ \mu\text{g/m}^3$, $4.9 \pm 6.0 \ \mu\text{g/m}^3$, $24.2 \pm 19.7 \ \mu\text{g/m}^3$, and $1.7 \pm 2.8 \ \text{ppm}$, in respective. The meteorological factor temperature was $26.7 \pm 3.7 \ ^{\circ}\text{C}$, and the RH was $59.6 \pm 8.4\%$. In terms of cardiovascular indicators, the mean \pm SD for each ln-transformed HRV parameter was as follows (Table A1): 4.4 ± 0.2 for ln HR, 3.4 ± 0.6 for ln r-MSSD, 4.1 ± 0.5 for ln SDNN, 1.8 ± 1.4 for ln pNN50 + 1, 6.5 ± 1.0 for ln LF, 5.4 ± 1.2 for ln HF, 7.8 ± 1.0 for ln TP, and 1.1 ± 0.8 for ln LF/HF. Table A2 presents the distribution of all participants' exposures in bus, MRT, and campus microenvironments. The average CO exposure on buses and MRT was $7.0 \ \text{vs}$. $2.4 \ \text{ppm}$, PM₁₀ of $64.1 \ \text{vs}$. $59.8 \ \mu\text{g/m}^3$, PM_{2.5–10} of $27.3 \ \text{vs}$. $17.4 \ \mu\text{g/m}^3$, PM_{2.5} of $36.8 \ \text{vs}$. $42.4 \ \mu\text{g/m}^3$, PM_{1-2.5} of $5.6 \ \text{vs}$. $9.2 \ \mu\text{g/m}^3$, PM₁ of $31.2 \ \text{vs}$. $33.2 \ \mu\text{g/m}^3$, temperature of $26.4 \ \text{vs}$. $26.2 \ ^{\circ}\text{C}$, and the RH of $57.3 \ \text{vs}$. 58.5%. As shown in the table, both of the exposures to PM and CO on bus or MRT were significantly higher than those in the campus microenvironment.

Table 1 summarizes the HRV distribution when participants were in the bus, MRT, and campus microenvironments. With the average of the HRV parameters of each participant on campus set as the control group, the averages of HRV parameters from different mass transits (bus or MRT) were compared. If the percentage changes for the parameters were positive, it means that the values of HRV parameters increased when the participants were on this vehicle; if the percentage was negative, it means that the values of HRV parameters decreased. In comparison with the participants' HRV variations on campus, the average ln HR variation on bus was 3.1%, with ln r-MSSD variation of -6.5%, ln SDNN variation of -5.8%, ln pNN50+1 variation of -9.3%, ln LF variation of -13.4%, ln HF variation of -17.3%, ln TP variation of -9.6%, and ln LF/HF variation of 22.1%. In addition, the average ln HR variation of -42.6%, ln LF variation of -6.5%, ln SDNN variation of -42.6%, ln LF variation of -6.7%, and ln LF/HF variation of -6.7%, and ln LF/HF variation of -6.7%, ln SDNN variation of -42.6%, ln LF variation of -6.8%, ln HF variation of -13.3%, ln TP variation of -42.6%. In brief, the HRV parameters (ln r-MSSD, ln SDNN, ln pNN50+1, ln LF, ln HF, ln TP) on bus and MRT were all lower than those on campus, except for ln HR and ln LF/HF.

	Campus	Bus	MRT	Bus-Camp #	MRT-Camp [#]
Variable	Mean ± SD (Range)				
ln HR	4.4 ± 0.2	4.5 ± 0.2	4.5 ± 0.1	3.1 ± 1.5	1.7 ± 1.5
(beats/min)	(3.6, 5.1)	(4.1, 5.0)	(4.2, 4.8)	(0.2, 5.6)	(-2.3, 3.6)
ln r MSSD (mc)	3.5 ± 0.6	3.2 ± 0.7	3.2 ± 0.6	-6.5 ± 15.2	-7.2 ± 10.5
III I-1035D (IIIS)	(2.0, 6.0)	(2.0, 6.0)	(2.0, 5.0)	(-21.6, 31.3)	(-19.4, 20.0)
In SDNN (ma)	4.1 ± 0.4	3.9 ± 0.5	3.9 ± 0.4	-5.8 ± 6.9	-5.5 ± 4.3
III SDININ (IIIS)	(2.0, 6.0)	(3.0, 5.0)	(3.0, 5.0)	(-17.8, 11.1)	(-10.8, 3.3)
ln pNN50 + 1	1.9 ± 1.4	1.1 ± 1.5	1.1 ± 1.4	-9.3 ± 152.5	-42.6 ± 80.9
(%)	(-1.8, 4.4)	(-1.8, 4.4)	(-1.8, 4.2)	(-139.6, 453.4)	(-150.4, 226.5)
1. IF (6.6 ± 1.0	5.7 ± 1.2	6.2 ± 0.9	-13.4 ± 7.1	-6.8 ± 4.7
In LF (ms ⁻)	(2.4, 13.8)	(0.9, 10.6)	(3.3, 9.7)	(-26.7, 7.1)	(-16.9, -0.8)
$\ln IIE (m n^2)$	5.5 ± 1.2	4.5 ± 1.4	4.8 ± 1.1	-17.3 ± 16.6	-13.3 ± 10.9
In FIF (ms ⁻)	(0.1, 10.9)	(1.3, 10.1)	(1.7, 9.4)	(-34.7, 27.2)	(-30.8, 9.8)
1 - TD(2 - 2)	7.8 ± 1.0	7.1 ± 1.1	7.3 ± 0.9	-9.6 ± 6.5	-6.7 ± 4.2
$\ln TP (ms^2)$	(0.0, 26.2)	(2.6, 11.5)	(4.7, 10.4)	(-23.4, 6.2)	(-13.0, 2.0)
In LE/LIE	1.1 ± 0.8	1.2 ± 0.9	1.4 ± 0.8	22.1 ± 77.7	42.2 ± 68.9
IN LF/HF	(-2.5, 4.1)	(-1.5, 3.2)	(-0.8, 3.2)	(-168.8, 217.1)	(-66.3, 257.5)

Table 1. Summary statistics of heart rate variability indices for the participants in different microenvironments.

[#] HRV change percent, %; Bus–Camp = [(Bus–Campus)/Campus] × 100%; MRT–Camp = [(MRT–Campus)/Campus] × 100%. HRV: heart rate variability. MRT: Mass Rapid Transit. HR: heart rate. r-MSSD: square root of the mean of the sum of the squares of differences between adjacent normal-to-normal (NN) intervals. SDNN: standard deviation of all NN intervals. pNN50: percentage of successive NN interval differences greater than 50 ms. LF: low-frequency power. HF: high-frequency power. TP: total power.

3.2. Impacts of PM and CO Exposures on HRV Indices

In the multi-pollutant model, the 5 min moving average concentrations of PM and CO both caused a percentage change in HR, and the change was statistically significant (Table 2). Specifically, an increase of 1 μ g/m³ of PM_{2.5-10} led to a significant increase in HR by 0.055% (95% confidence interval: 0.046, 0.064); an increase of 1 μ g/m³ of PM_{1-2.5} led to a significant decrease in HR by 0.137% (-0.248, -0.026); an increase of 1 μ g/m³ of PM₁ led to a significant increase in HR by 0.106% (0.063, 0.149); and an increase of 1 ppm of CO led to a significant decrease in HR by 0.435% (-0.670, -0.201). The percentage change of SDNN was statistically significant for each increase of 1 μ g/m³ of PM_{1-2.5}, and PM₁, which were 0.082% (0.032, 0.131), -0.513% (-0.943, -0.081), and 0.209% (0.081, 0.338), respectively. In addition, PM_{2.5-10} significantly reduced the percentage change of HF by 0.180% (-0.304, -0.056), and the percentage change of LF/HF increased significantly by 0.155% (0.049, 0.261). Overall, PM_{2.5-10} and PM₁ both increased the percentage change of HR, and CO decreased the percentage change of HR. In terms of time domain HRV indices, PM_{2.5-10} and PM₁ both significantly increased the percentage change of SDNN. For the frequency domain parameters, PM_{2.5-10} caused a significant decrease in the percentage change of HF, while making the percentage change of LF/HF rise significant parameters.

Variable		Multi-Pollutant Models *					
	PM _{2.5-10}	PM _{1-2.5}	PM ₁	СО			
LID	0.055 ⁺	-0.137 ⁺	0.106 ⁺	-0.435 ⁺			
ПК	(0.046, 0.064)	(-0.248, -0.026)	(0.063, 0.149)	(-0.670, -0.201)			
* MCCD	0.036	-0.506	0.032	-0.359			
r-M55D	(-0.018, 0.090)	(-1.012, 0.003)	(-0.132, 0.196)	(-1.515, 0.811)			
	0.082 +	-0.513 ⁺	0.209 ⁺	0.202			
SDNN	(0.032, 0.131)	(-0.943, -0.081)	(0.081, 0.338)	(-0.783, 1.197)			
mNINE0+1	-0.047	-0.262	-0.128	0.294			
pinin50+1	(-0.121, 0.026)	(-1.088, 0.571)	(-0.435, 0.181)	(-1.488, 2.108)			
τr	-0.086	0.664	-0.060	1.576			
LF	(-0.201, 0.029)	(-0.320, 1.656)	(-0.338, 0.218)	(-0.618, 3.819)			
LIF	-0.180 ⁺	0.415	-0.034	1.473			
HF	(-0.304, -0.056)	(-0.704, 1.546)	(-0.374, 0.307)	(-1.089, 4.102)			
ТD	0.034	-0.074	0.189	1.119			
IP	(-0.077, 0.145)	(-1.018, 0.880)	(-0.078, 0.456)	(-1.018, 3.301)			
I E/LIE	0.155 ⁺	0.082	0.032	0.176			
LF/HF	(0.049, 0.261)	(-0.841, 1.014)	(-0.233, 0.298)	(-1.840, 2.234)			

Table 2. Impacts of personal particulate matter (PM) and carbon monoxide (CO) exposures on HRV indices in multi-pollutant models.

* All models are 5 min moving average of pollutants; coefficients are expressed as % change (95 confidence interval) in HRV for increase in pollutants exposures in models adjusting for subject, sex, BMI (body mass index), location, temperature, RH (relative humidity), day of week. [†] Statistical significance in bold (p < 0.05).

3.3. Short-Term Exposure Impacts on HRV Indices in Different Time Frames

As shown in Table 3, in terms of the moving average concentrations of PM, $PM_{2.5-10}$ caused a positive percentage change of HR regardless of the length of time periods. It was found that with the increase of the moving average time, $PM_{2.5-10}$ had a significant increase in the percentage change (0.055%, 0.106%, and 0.169%, respectively). In terms of moving average concentration of $PM_{1-2.5}$, as the moving average time increased, percentage change significantly decreased (-0.137%, -0.551%, and -0.994%, respectively). The 60 min moving average concentration of PM_1 significantly increased the percentage change of HR by 0.125%.

Additionally, as the moving average time increased, the moving average concentration of $PM_{2.5-10}$ had a slight decrease in the percentage change of r-MSSD. On the contrary, the moving average concentration of $PM_{1-2.5}$ showed a gradual upward trend along with the increase in the moving average time. The 60 min moving average concentration of CO significantly reduced the percentage change of r-MSSD by 2.548%.

The analysis of the percentage change of SDNN showed that as the average time increased, the moving average concentration of $PM_{2.5-10}$ led to a significant decrease in the percentage change of SDNN, while $PM_{1-2.5}$ led to a significant increase of the change of this parameter. The 60 min moving average concentrations of $PM_{2.5-10}$ and $PM_{1-2.5}$ resulted in a significant decrease of 0.194% and significant increase of 1.139%, respectively. The moving average concentration of CO decreased the percentage change of SDNN with the increase in moving average time periods, where the 60 min average moving concentration of CO resulted in a significant decrease of 1.844%.

In terms of pNN50+1, the moving average concentration of $PM_{2.5-10}$ led to a slight decrease in the percentage change of pNN50+1 as the moving average time increased. On the other hand, the moving average concentration of $PM_{2.5-10}$ increased the percentage change of pNN50+1 as the moving average time increased. The 60 min moving average concentration of $PM_{2.5-10}$ significantly reduced pNN50+1 by 0.472%, while that of $PM_{1-2.5}$ led to an increase of pNN50+1 by 2.702%.

	Moving	Multi-Pollutant Models *					
Variable	Average	PM _{2.5-10}	PM _{1-2.5}	PM ₁	СО		
	5 min	0.055 ⁺ (0.046, 0.064)	-0.137 ⁺ (-0.248, -0.026)	0.106 ⁺ (0.063, 0.149)	-0.435 ⁺ (-0.670, -0.201)		
HR	30 min	0.106 ⁺ (0.072, 0.141)	-0.551 ⁺ (-0.822, -0.280)	0.030 (-0.043, 0.104)	-0.130 (-0.655, 0.397)		
	60 min	0.169 ⁺ (0.115, 0.222)	-0.994 ⁺ (-1.377, -0.608)	0.125 ⁺ (0.045, 0.204)	-0.222 (-0.993, 0.555)		
	5 min	0.036 (-0.018, 0.090)	-0.506 (-1.012, 0.003)	0.032 (-0.132, 0.196)	-0.359 (-1.515, 0.811)		
r-MSSD	30 min	-0.084 (-0.208, 0.041)	0.454 (-0.483, 1.400)	-0.047 (-0.261, 0.168)	-0.348 (-2.090, 1.425)		
	60 min	-0.155 (-0.312, 0.003)	1.038 (-0.120, 2.210)	-0.032 (-0.274, 0.210)	-2.548 ⁺ (-4.676, -0.374)		
	5 min	0.082 ⁺ (0.032, 0.131)	-0.513 ⁺ (-0.943, -0.081)	0.209 ⁺ (0.081, 0.338)	0.202 (-0.783, 1.197)		
SDNN	30 min	-0.083 (-0.179, 0.014)	0.480 (-0.240, 1.206)	0.102 (-0.059, 0.263)	-0.745 (-2.062, 0.590)		
	60 min	-0.194 ⁺ (-0.313, -0.075)	1.139 ⁺ (0.260, 2.026)	0.104 (-0.079, 0.286)	-1.844 ⁺ (-3.424, -0.238)		
pNN50 + 1	5 min	-0.047 (-0.121, 0.026)	-0.262 (-1.088, 0.571)	-0.128 (-0.435, 0.181)	0.294 (-1.488, 2.108)		
	30 min	-0.210 (-0.449, 0.031)	0.757 (-1.100, 2.648)	-0.027 (-0.499, 0.447)	1.621 (-1.944, 5.316)		
	60 min	-0.472 ⁺ (-0.817, -0.126)	2.702 ⁺ (0.156, 5.314)	-0.352 (-0.871, 0.168)	-2.521 (-7.213, 2.409)		

Table 3. Short-term exposure impacts on time domain HRV indices in multi-pollutant models.

* Coefficients are expressed as % change (95 confidence interval) in HRV for one unit increase in pollutants exposures in models adjusting for subject, sex, BMI, location, temperature, RH, day of week. [†] Statistical significance in bold (p < 0.05).

With regard to frequency domain HRV parameters, as shown in Table 4, $PM_{2.5-10}$ made the percentage change of LF slightly decrease as the moving average time period lengthened; 30 to 60 min moving average concentrations of $PM_{2.5-10}$ led LF's percentage change to decrease significantly by 0.459% and 0.598%. As the moving average time increased, the moving average concentration of $PM_{1-2.5}$ caused the percentage change of LF to increase gradually, and the 30 to 60 min average moving concentration of $PM_{1-2.5}$ led the percentage change of LF to increase significantly by 2.779% and 3.748%.

As the moving average time increased, the moving average concentration of $PM_{2.5-10}$ caused a slight decrease in the percentage change of HF; the percentage changes of HF caused by the 5 to 60 min moving average concentration were significantly decreased by 0.180%, 0.625%, and 0.845%. Conversely, as the moving average time increased, the moving average concentration of $PM_{1-2.5}$ caused the percentage change of HF to increase gradually. The 30 to 60 min moving average concentration resulted in a significant increase in HF by 3.539% and 5.260%.

With the increase of the moving average time period, it was found that the $PM_{2.5-10}$ and CO moving average concentrations caused the percentage change of TP to decrease gradually, and the 60 min average moving concentrations of $PM_{2.5-10}$ and CO led to the greatest percentage change of TP, which decreased significantly by 0.484% and 3.366%, respectively. Nonetheless, the moving average concentration of $PM_{1-2.5}$ caused the percentage change of TP to increase gradually along with the elongation of the moving average time period, and the percentage change of TP caused by the 30 and 60 min moving average concentrations increased significantly by 1.890% and 2.846%.

	Moving	Multi-Pollutant Models *				
variable	Average	PM _{2.5-10}	PM _{1-2.5}	PM ₁	СО	
	5 min	-0.086	0.664	-0.060	1.576	
	5 mm	(-0.201, 0.029)	(-0.320, 1.656)	(-0.338, 0.218)	(-0.618, 3.819)	
IF	30 min	-0.459 ⁺	2.779 ⁺	-0.147	-1.849	
LI	50 11111	(-0.666, -0.251)	(1.198, 4.385)	(-0.489, 0.196)	(-4.544, 0.932)	
	60 min	-0.598 ⁺	3.748 ⁺	-0.173	-2.747	
	00 11111	(-0.852, -0.343)	(1.834, 5.698)	(-0.559, 0.215)	(-5.914, 0.526)	
	E	-0.180 ⁺	0.415	-0.034	1.473	
	5 min	(-0.304, -0.056)	(-0.704, 1.546)	(-0.374, 0.307)	(-1.089, 4.102)	
UE	20	-0.625 ⁺	3.539 ⁺	-0.315	-0.161	
ПГ	50 mm	(-0.880, -0.370)	(1.566, 5.549)	(-0.745, 0.117)	(-3.686, 3.494)	
	(0	-0.845 ⁺	5.260 ⁺	-0.449	-3.012	
	60 min	(-1.162, -0.527)	(2.827, 7.751)	(-0.936, 0.040)	(-7.181, 1.345)	
	- ·	0.034	-0.074	0.189	1.119	
	5 min	(-0.077, 0.145)	(-1.018, 0.880)	(-0.078, 0.456)	(-1.018, 3.301)	
тр	20	-0.319 ⁺	1.890 ⁺	0.056	-1.568	
11	50 mm	(-0.516, -0.121)	(0.399, 3.403)	(-0.270, 0.382)	(-4.220, 1.157)	
	60 min	-0.484 ⁺	2.846 ⁺	0.101	-3.366 ⁺	
	60 mm	(-0.725, -0.242)	(1.042, 4.682)	(-0.268, 0.471)	(-6.480, -0.149)	
	- .	0.155 ⁺	0.082	0.032	0.176	
LF/HF	5 min	(0.049, 0.261)	(-0.841, 1.014)	(-0.233, 0.298)	(-1.840, 2.234)	
	20	0.235 ⁺	-0.730	0.208	-2.235	
	30 min	(0.033, 0.439)	(-2.216, 0.779)	(-0.125, 0.543)	(-4.816, 0.415)	
	(0 min	0.284 +	-1.286	0.288	-1.293	
	60 min	(0.034, 0.535)	(-3.066, 0.526)	(-0.091, 0.668)	(-4.395, 1.911)	

Table 4. Short-term exposure impacts on frequency domain HRV indices in multi-pollutant models.

* Coefficients are expressed as % change (95 confidence interval) in HRV for one unit increase in pollutants exposures in models adjusting for subject, sex, BMI, location, temperature, RH, day of week. [†] Statistical significance in bold (p < 0.05).

Furthermore, as the moving average time increased, the moving average concentration of $PM_{2.5-10}$ caused the percentage change in LF/HF to significantly increase by 0.155%, 0.235%, and 0.284%, respectively. The moving average concentration of $PM_{1-2.5}$ tended to decrease gradually with the increase of the moving average time. As the moving average time increased, the moving average concentration of PM_1 caused a slight increase in the percentage change of LF/HF, yet this trend is not statistically significant.

To sum up, current results showed that $PM_{2.5-10}$ caused a short-term decrease in the percentage changes of the r-MSSD, SDNN, pNN50+1, LF, HF, and TP with the increase of the moving average time in the multi-pollutant models. $PM_{1-2.5}$ led to a short-term gradual increase in the percentage changes of r-MSSD, SDNN, pNN50+1, LF, HF, and TP along with the increase of moving average time periods. As the moving average time increased, PM_1 caused a short-term slight decrease in the percentage changes of r-MSSD, pNN50+1, LF, and HF, whereas CO caused a short-term gradual decrease in the percentage changes of r-MSSD, SDNN, LF, HF, and TP. The majority of the impacts caused by $PM_{2.5-10}$ and $PM_{1-2.5}$ were statistically significant, while the impacts of PM_1 and CO did not show statistical significance most of the time.

3.4. Impact of Personal Exposures in Public Transportation Microenvironments on HRV Indices

As shown in Table 5, compared with the on-campus microenvironment, exposures to PM_1 on bus increased the percentage change of HR (6.98%), which was slightly larger than other pollutants. $PM_{2.5-10}$ reduced the percentage change of LF and HF (-58.60% and -59.81%, respectively), which indicated a slightly greater tendency than other pollutants. $PM_{1-2.5}$ reduced the percentage change of r-MSSD, SDNN, pNN50+1, and TP (-20.49%, -25.44%, -27.61%, and -53.48%, respectively), which was slightly larger than other pollutants. The degree to which CO caused the percentage change of LF/HF (4.82%) tended to be slightly larger than PM with different sizes. On the other hand, compared with the on-campus microenvironment, exposures to CO on MRT increased the percentage change of HR (3.25%), which was greater than size-fractionated PM. $PM_{2.5-10}$ reduced the percentage changes of r-MSSD, SDNN, pNN50+1, LF, HF, and TP (-22.26%, -25.19%, -29.07%, -46.90%, -55.72%, and -47.84%, respectively), which is slightly larger than other pollutants. The degree to which CO led to the percentage changes of LF/HF (19.81%) had a tendency to be slightly larger than size-fractionated PM. In general, it can be found that compared with the on-campus microenvironment, when participants were exposed to PM of various particle sizes and CO on bus, the percentage change of HRV had a tendency to decrease, while the percentage change of HR and LF/HF had a trend to increase. When participants were exposed to $PM_{2.5-10}$ on MRT, compared with the on-campus microenvironment, the percentage changes in time domain and frequency domain HRV indices (except LF/HF) were greater than other pollutants. The extent to which CO increased the percentage changes of HR was greater than that of PM with different sizes.

	Multi-Pollutant Models *							
Variable	PM ₂	.5–10	PM	1–2.5	PN	M ₁	C	0
-	Bus	MRT	Bus	MRT	Bus	MRT	Bus	MRT
HR	6.93	3.13	6.74	3.18	6.98	3.15	6.44	3.25
r-MSSD	-19.94	-22.26	-20.49	-22.03	-19.95	-22.21	-20.34	-21.69
SDNN	-24.84	-25.19	-25.44	-24.99	-24.72	-25.15	-24.72	-24.71
pNN50+1	-27.39	-29.07	-27.61	-28.68	-27.47	-28.95	-27.05	-28.19
LF	-58.60	-46.90	-57.85	-46.46	-58.58	-46.82	-56.94	-45.84
HF	-59.81	-55.72	-59.22	-55.21	-59.67	-55.61	-58.16	-54.47
TP	-53.37	-47.84	-53.48	-47.42	-53.22	-47.77	-52.29	-46.81
LF/HF	4.80	18.82	4.72	19.24	4.67	18.90	4.82	19.81

Table 5. Impact of microenvironmental personal exposures in public transportation on HRV indices.

* All models are 5 min moving average of pollutants; coefficients are expressed as % change in HRV for one unit increase in pollutants exposures in models adjusting for subject, sex, BMI, temperature, RH, day of week.

4. Discussion

This study examined the effects of the exposure to pollutants on HRV with 20 healthy adults as participants who commuted via buses and MRTs. The results showed that when the participants took the bus, they were exposed to higher CO concentrations than in MRT. This finding is in accord with Wu's previous study, which found that the CO concentration in the bus fell between 1.5 and 5.4 ppm, and the CO concentration in MRT fell between 0.5 and 2.6 ppm [34]. Actually, when bus passengers get on and off the vehicle, CO enters the cabin by diffusion. CO in MRT is not generated by the operation of the equipment in MRT, but is externally diluted by the atmosphere and then diffused into the MRT. Therefore, the CO concentration in bus is higher than that in MRT. Besides, the concentrations of PM_{2.5-10} and PM₁₀ in bus are higher than those in MRT in the current research. Specifically, $PM_{2.5-10}$ concentration fell between 0.6 and 470.5 μ g/m³ vs. 0.2 to 305.4 μ g/m³, and PM_{10} concentration fell between 11.0 and 505.4 $\mu g/m^3$ vs. 10.6 to 363.6 $\mu g/m^3$. The results also tally with Wu's, which reported that $PM_{2.5-10}$ concentration in bus and MRT was 22.5–46.1 µg/m³ vs. 20.4–32.6 µg/m³, and PM₁₀ concentration was 60.5–75.0 µg/m³ vs. 58.2–76.1 µg/m³. Also, Wu found that the concentration of PM_{2.5-10} increased after the bus opened its door, therefore pollutant exposures in bus are more susceptible to road traffic pollutants [34]. As a result, the participants were exposed to higher concentrations of carbon monoxide and coarse particles in bus than in MRT.

Results of the GAMM analysis were used to observe the influence on participants' HRV when they were exposed to pollutants in bus and MRT. It was found that compared with the on-campus situation, the 5 min moving average concentration exposure to PM and CO in bus or MRT showed a downward trend in HRV (except LF/HF), as shown in Table 3. This finding is in line with the results of Adar et al. [16]. Adar et al. observed the relationship between exposure to traffic particle concentrations and

24 h HRV in 44 elderly people with an age of over 60. Their results showed that viewed from the perspective of the vehicle that the participants took, along with an increase of an interquartile range (IQR) of 10 μ g/m³, the 5 min average concentration of PM_{2.5} in buses caused HF to decrease by 11%. The current study found that along with each increase of 1 μ g/m³ in PM_{1-2.5} to which participants were exposed on the bus, the percentage change of HF for them decreased by 57.76%, and PM₁ caused the percentage changes of HF in healthy adults to decrease by 58.15%. The reason why the percentage change of HF in the current study differs from that in Adar et al. may be the difference in participants' age and the disease they got, as well as in the time when participants took buses. Adar et al. designed one hour for a one-way bus ride, but this research was about 20 min. Furthermore, it is also possible that the HRV variation is greater in this study due to the better adaptability of the young population to the external environmental changes [35].

In terms of short-term effects caused by particles, this study found that $PM_{2.5-10}$ caused a slight decrease in the percentage changes of time domain parameters (r-MSSD, SDNN, and pNN50+1) and frequency domain parameters (LF, HF, and TP) with an increase in moving average time periods. This effect can last as long as 60 min (Tables 2 and 3). Yeatts et al. found that every increase of 1 µg/m³ in $PM_{2.5-10}$ caused the SDNN of adults with asthma to decrease by 3.36% in 24 h and their HF to decrease by 0.46% [27]. In addition, Lipset et al. studied the relationship between exposure to $PM_{2.5-10}$ and HRV for the elderly, and found that $PM_{2.5-10}$ caused a decline in the SDNN [28]. Domestic researchers, such as Chang et al., also obtained results in line with this study. Every increase of 1 µg/m³ in the 6 h moving average concentration exposure of $PM_{2.5-10}$ caused the r-MSSD and SDNN for the elderly to decline by 4.27% and 1.43%, respectively [26]. Therefore, this study infers that $PM_{2.5-10}$ not only reduces the HRV of the elderly, but also affects healthy adults [36]. In addition, this effect may last more than 60 min.

In addition, this study found that the moving average concentration of $PM_{1-2.5}$ caused the percentage change of healthy adults' HR and LF/HF to decrease with the increase of the moving average time, whereas it caused the percentage change of r-MSSD, pNN50+1 and HF to increase with the increase of the moving average time. This finding is different from the effect of $PM_{2.5-10}$ on HRV. Wu et al. found that $PM_{1-2.5}$ increased the right cardio-ankle vascular index (CAVI), an arterial stiffness index, results of which once again highlights the importance of having size-fractionated PM data; otherwise, we would be unable to identify the particulate matter effects from the commonly used $PM_{2.5}$ and PM_{10} variables [37]. However, there is still limited discussion on the relationship between the fine particle size of $PM_{1-2.5}$ and the influence of HRV, which needs further research.

This study found that PM_1 caused a slight decrease in the percentage change of LF and HF in the multi-pollution model. This finding is different from Chuang's [29]. Chuang et al. found that each increase of IQR (28.3 µg/m³) in the 1 to 3 h moving average concentration of $PM_{0.3-1.0}$ resulted in a decrease of 8.25% in r-MSSD of those who had coronary artery diseases, a decrease of 4.88% in their SDNN, a decrease of 3.83% in LF, and a decrease of 5.28% in HF. For patients with hypertension, each increase of IQR (27.2 µg/m³) in 1 to 3 h moving average concentration of $PM_{0.3-1.0}$ resulted in a 2.73% decrease in r-MSSD, a 1.49% decrease in SDNN, a 1.86% decrease in LF, and a 2.84% decrease in HF. Chuang et al. speculated that the effects of $PM_{0.3-1.0}$ on health may be caused by the differences in the chemical and biological components of the particles, rather than the difference in the particle size. In fact, due to the different physical and chemical properties, fine particles can penetrate deep into lung tissues and the fine particles with 5 to 10 nm can be transported to the whole body through blood vessels, causing an inflammatory reaction or leading to an increasing heart rate by stimulating the sympathetic or parasympathetic nerves. As the heart rate increases, the relative heart rate variability becomes smaller, resulting in direct cardiovascular damage [38].

On the other hand, the current study found that CO decreased healthy adults' percentage change in SDN, LF, HF, and TP over time, which is in line with the research results of Schwartz et al. [39]. Each increase of IQR (0.45 ppm) in a 24 h average concentration of CO would reduce the SDNN of the elderly by 2.6%. The results of this study are also similar to those of Timonen et al. [40]. When CO exposure concentration increased by 1 mg/m³, the elderly's SDNN decreased by 5.69%. Riojas-Rodríguez et al.

found that every increase of 1 ppm in CO concentration caused the ln LF of patients with the ischemic heart disease to decrease by 0.024 [41]. An increase in particles and CO leads to subtle changes in respiratory regulation, resulting in HRV changes [42]. This study found that with the elongation of the moving average time periods, CO not only reduced healthy adults' percentage change of SDNN, but also decreased the percentage changes of LF, HF, and TP in a short term. Consequently, this study speculates that exposure to CO also causes the HRV of the elderly and healthy adults to decline, which may exert a negative impact on their health.

The strength of this panel study was the continuous personal measurement data on a small time scale, which could directly be related to HRV parameters. Therefore, instant associations could be captured, which was not possible when data from a stationary monitoring site had been used. The collection of individual-level outcome data also means that hypothesis testing can provide strong evidence of associations at that level. Each subject acts as his (or her) own control, and only covariates that vary across time within an individual need be considered by the analysis [21]. A disadvantage of this study was the small number of participants. However, a large number of repeated 5 min monitoring segments were available for each participant. Still, larger sample sizes than those available in this study would be required to definitively conclude the effects of exposures to air pollutants. In addition, we could not exclude the confounding effect of respiration on the HRV indices because the association of the participants' breathing patterns with vagal cardiac outflow was not measured in the current study [43]. Physical activity and stress might alter breathing patterns and heart rate and then influence HRV measurements in the walking mode [24]. Finally, researchers have pointed out that noise may be one of the factors that potentially interfere with the autonomic nervous system, which in turn affects the response of HRV [30]. Consequently, it is suggested that noise data can be added to future studies.

5. Conclusions

In this study, continuous personal measurement data were collected to investigate the influence of size-fractionated particles and CO on the HRV indicators of healthy adults at 5 min, 30 min, and 60 min time periods and the HRV impacts of in-cabin exposure to air pollutants by taking public transportation (bus and MRT). Results of the GAMM analysis showed that both $PM_{2.5-10}$ and PM_1 caused the percentage change of HR to increase significantly, and the percentage change of SDNN, a time domain HRV parameter, to increase significantly; $PM_{2.5-10}$ caused the percentage change of HF, a frequency domain HRV parameter, to decrease significantly and the percentage change of LF/HF to increase significantly; CO caused the percentage change of HR to decrease significantly, and the percentage changes of frequency domain HRV parameters to show an upward trend, but did not reach statistical significance.

This study also found that, with the elongation of moving average time periods, the HRV of healthy adults who were exposed to size-fractionated particle were affected; $PM_{2.5-10}$ caused a short-term slight decrease in the percentage change of r-MSSD, SDNN, pNN50+1 LF, HF, and TP. Moreover, when 60 min moving average concentration of $PM_{2.5-10}$ increased, the percentage of pNN50+1, LF, HF, and TP decreased significantly; $PM_{1-2.5}$ caused a short-term gradual decrease in the percentage change of HR and LF/HF, but a short-term gradual increase in the percentage changes of r-MSSD, pNN50+1, and HF; CO caused a short-term gradual decrease in the percentage of SDNN, LF, HF, and TP, and the increase in 60 min moving average concentration of CO caused the largest decrease in the percentage changes of SDNN and TP with statistical significance.

Compared with the campus, PM_1 exposure on bus caused a slightly greater increase in the percentage change of HR than other pollutants whereas $PM_{2.5-10}$ caused a slightly greater decrease in the percentage change of LF and HF than other pollutants. $PM_{1-2.5}$ caused a slightly greater decrease in the percentage change of r-MSSD, SDNN, pNN50+1, and TP than other pollutants, while CO caused a slightly greater increase in the percentage change of LF/HF than PM. Moreover, $PM_{2.5-10}$ exposure on MRT tended to cause a greater decrease in the percentage change in time domain and frequency

domain HRV indices (except LF/HF) than other pollutants, whereas CO tended to cause a greater increase in the percentage change of HR than PM.

In summary, particles with different sizes cause different variations of the HRV indices. Among them, $PM_{2.5-10}$ results in much variation of the HRV parameters. Therefore, exposure to $PM_{2.5-10}$ has a slightly stronger effect on the autonomic nervous system of the heart of healthy adults, while the effect of finer-sized particles on the HRV remains to be confirmed by subsequent studies.

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Appendix A

Table A1. Summary statistics for the participants	air pollution exposures, heart rate variability and
meteorological conditions using a 5-min mean.	

Variable	Ν	$Mean \pm SD$	Minimum	Maximum
ln HR ¹ (beats/min)	5613	4.4 ± 0.2	3.6	5.1
Time domain HRV 1				
ln r-MSSD(ms)	5609	3.4 ± 0.6	2.0	6.0
ln SDNN(ms)	5609	4.1 ± 0.5	2.0	6.0
ln pNN50+1 (%)	5609	1.8 ± 1.4	-1.8	4.4
Frequency domain HRV ¹				
ln LF (ms ²)	5599	6.5 ± 1.0	0.9	13.8
ln HF (ms²)	5599	5.4 ± 1.2	0.1	10.9
ln TP(ms ²)	5599	7.8 ± 1.0	0.0	26.2
ln LF/HF	5599	1.1 ± 0.8	-2.5	4.1
Personal exposures ²				
CO (ppm)	7877	1.7 ± 2.8	0.0	22.8
PM ₁₀ (μg/m ³)	7926	45.4 ± 50.6	2.4	1131.3
PM _{2.5-10} (µg/m ³)	7926	16.3 ± 37.1	0.2	923.8
$PM_{2.5} (\mu g/m^3)$	7926	29.0 ± 23.8	1.9	233.5
PM _{1-2.5} (μg/m ³)	7926	4.9 ± 6.0	0.0	149.3
PM ₁ (μg/m ³)	7926	24.2 ± 19.7	1.1	206.2
Temperature ² (°C)	7956	26.7 ± 3.7	12.8	34.9
RH ² (%)	7955	59.6 ± 8.4	33.6	90.9

¹ The values do not include the measurements of sleep. ² The values include the measurements of sleep.

	Campus	MRT	Bus
Personal Exposures	Mean ± SD (Range)	Mean ± SD ⁺ (Range)	Mean ± SD ⁺ (Range)
CO(nnm)	1.5 ± 2.6	2.4 ± 3.0 *	7.0 ± 4.8 *
CO (ppill)	(0.0, 25.0)	(0.0, 14.0)	(0.0, 23.0)
$\mathbf{D}\mathbf{M} = (1, 2, 2, 2, 3)$	43.1 ± 53.6	59.8 ± 40.2 *	64.1 ± 50.9 *
$PM_{10} (\mu g/m^2)$	(1.1, 1231.3)	(10.6, 363.6)	(11.0, 505.4)
$\mathbf{D}\mathbf{M} = (-1)^3$	15.2 ± 40.8	17.4 ± 27.1 *	27.3 ± 44.3 *
$PM_{2.5-10} (\mu g/m^{\circ})$	(0.0, 1152.7)	(0.2, 305.4)	(0.6, 470.5)
\mathbf{D}	27.9 ± 23.7	42.4 ± 23.2 *	36.8 ± 22.8 *
$PM_{2.5} (\mu g/m^2)$	(1.1, 382.9)	(4.4, 197.2)	(6.9, 189.4)
\mathbf{D}	4.6 ± 6.2	9.2 ± 7.1 *	5.6 ± 4.1 *
$PM_{1-2.5} (\mu g/m^{\circ})$	(0.0, 290.1)	(0.3, 41.8)	(0.2, 32.9)
$\mathbf{D}(\mathbf{A}_{1}, \mathbf{A}_{2}, \mathbf{A}_{3})$	23.3 ± 19.6	33.2 ± 18.6 *	31.2 ± 20.2 *
$PM_1 (\mu g/m^2)$	(1.0, 211.4)	(4.1, 189.8)	(5.8, 183.9)
Torrest and have (°C)	26.8 ± 3.7	26.2 ± 2.6 *	26.4 ± 4.4
Temperature (°C)	(13.2, 34.8)	(19.9, 33.6)	(13.1, 35.1)
$\mathbf{D}_{\mathbf{r}}$ let in a let $\mathbf{r}_{\mathbf{r}}$ (0/)	59.6 ± 8.4	58.5 ± 6.0 *	57.3 ± 13.4 *
Relative numidity (%)	(33.3, 95.4)	(36.2, 75.5)	(32.2, 90.8)

Table A2.Personal exposures to air pollution and meteorological conditions indifferent microenvironments.

⁺ Mann-Whitney test, MRT versus Campus, Bus versus Campus. * Statistical significance in bold (p < 0.05).

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