

# Synergistic Effects of 1 h Post-Load Plasma Glucose and Smoking on Arterial Stiffness in Apparently Healthy Men: A Cross-sectional Study

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**Aim:** One-hour post-load plasma glucose (1h-PG) during an oral glucose tolerance test and smoking are associated with arterial stiffness. However, it remains unknown whether there are synergistic effects of these two factors on arterial stiffness. This study aimed to investigate the interaction between 1h-PG and smoking in relation to brachial–ankle pulse wave velocity (baPWV) in young men with normal glucose tolerance (NGT).

**Methods:** The study included 25-, 30-, 35-, 40-, and 45-year-old non-industrial male workers ( $n=2189$ ) who underwent a detailed health check-up. Normotensive participants with NGT and taking no medication were included.

**Results:** A univariate linear regression analysis showed that 1h-PG correlated with baPWV ( $r=0.13$ ,  $p<0.001$ ), but the correlation was not significant in the multivariate analysis ( $\beta=0.02$ ,  $p=0.24$ ). However, we found a significant interaction between 1h-PG levels and smoking status in relation to baPWV ( $p=0.048$ ). Therefore, further analyses were conducted in nonsmokers and smokers. A multivariate linear regression analysis revealed that 1h-PG significantly correlated with baPWV in smokers ( $\beta=0.11$ ,  $p=0.02$ ), but not in nonsmokers ( $\beta=0.01$ ,  $p=0.79$ ). The correlation remained significant even after adjustment for the number of cigarettes smoked per day ( $\beta=0.096$ ,  $p=0.048$ ) or the Brinkman index ( $\beta=0.097$ ,  $p=0.043$ ).

**Conclusion:** A significant interaction between 1h-PG and smoking in relation to baPWV was found in apparently healthy men younger than 50 years old.

**Key words:** Arterial stiffness, Smoking, Brachial–ankle pulse wave velocity, Oral glucose tolerance test, 1 h post-load plasma glucose

## Introduction

Smoking and diabetes mellitus (DM) are established risk factors of cardiovascular diseases<sup>1, 2</sup>. Accumulating evidence suggests that arterial stiffness, which is an independent predictor for cardiovascular events and mortality<sup>3, 4</sup>, is a common underlying pathophysiological mechanism related to the detrimental effects of smoking and DM<sup>5</sup>. Chronic cigarette smoking is associated with increased arterial stiffness even in young healthy people<sup>6, 7</sup>. Increased arterial stiffness also occurs in people with DM and even in people with impaired

glucose tolerance<sup>8, 9</sup> which is a risk factor in cardiovascular disease and mortality<sup>10</sup>.

Normal glucose tolerance (NGT) is considered to indicate a low cardiovascular risk state. However, several studies have reported that elevated 1 h post-load plasma glucose (1h-PG) levels obtained during an oral glucose tolerance test in people with NGT are associated with subclinical cardiovascular organ damage evidenced by left ventricular hypertrophy<sup>11</sup>, carotid atherosclerosis<sup>12</sup>, and arterial stiffness<sup>13-15</sup>. In our previous study of 25- to 55-year-old healthy participants, there was a significant interaction between 1h-PG and

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age in relation to arterial stiffness assessed by brachial–ankle pulse wave velocity (baPWV)<sup>16</sup> in men. The association between 1h-PG and baPWV became evident with aging<sup>15</sup>. In fact, a significant association was found only in middle-aged men older than 50 years. No significant association was found in women in any of the age groups. One possible explanation for this finding is that people with existing stiff vessels, middle-aged men, in this case, are more susceptible to vascular damage caused by postprandial hyperglycemia than those characterized by soft and healthy arteries such as young men and young to middle-aged women<sup>17</sup>. Thus, we hypothesized that 1h-PG would be associated with increased PWV if there were some factors increasing arterial stiffness or having synergistic effects on the progress of arterial stiffness even in young people. In this study, we focused on smoking as one such factor because smoking is associated with increased arterial stiffness and is an important albeit avoidable cause of cardiovascular diseases.

## Aim

This study aimed to investigate the synergistic effects of 1h-PG and smoking on arterial stiffness in men younger than 50 years old. The association between 1h-PG and baPWV was examined in nonsmokers and smokers separately.

## Participants and Methods

### Study Population

Study participants were non-industrial workers in central and branch offices in Japan. They underwent a detailed health check-up every 5 years from 25 to 55 years of age at the Chiba Foundation for Health Promotion & Disease Prevention. In this study, we focused on men younger than 50 years old in whom 1h-PG did not correlate with the baPWV values obtained in our previous study<sup>15</sup>. We excluded women because of the low frequency of smoking. Thus, the participants of this study were 25-, 30-, 35-, 40-, and 45-year-old male workers ( $n=3336$ ) who had a check-up between April 2011 and March 2016. Among them, people with NGT, as defined by the American Diabetes Association criteria, were included<sup>18</sup>. These criteria included fasting plasma glucose concentrations  $<100$  mg/dL, 2 h post-load plasma glucose (2h-PG) concentrations  $<140$  mmHg, and HbA1c levels  $<5.7\%$ . We excluded participants with cardiovascular diseases, hypertension (systolic blood pressure (SBP)  $\geq 140$  mmHg or diastolic blood pressure (DBP)  $\geq 90$  mmHg), those taking any medications, those with an abnormal ankle–brachial index (ABI) ( $ABI \leq 1.0$ ,  $ABI \geq 1.3$ ), those with an abnormal

white blood cell count ( $WBC < 3.0$ ,  $WBC > 11.0 \times 10^9/L$ ), and those with insufficient data on self-reported smoking status including the number of cigarettes smoked per day and the number of years of smoking. The final sample consisted of 2189 participants (1797 nonsmokers and 392 smokers). All the participants were asked not to smoke from midnight prior to the check-up day. The institutional ethics committee approved the study protocol. Written informed consent was obtained from each participant.

### Measurements

The baPWV was measured after a rest period of  $\geq 5$  min in a supine position using a commercially available device (form PWV/ABI, Omron Colin, Tokyo, Japan) that provides simultaneous measurements of bilateral brachial and ankle blood pressure and ABI<sup>16</sup>. The mean bilateral baPWV values were used as the baPWV value. ABI was defined as the ratio of the ankle SBP to the brachial SBP.

Venous blood samples after an overnight fast (0 min) and at 60 and 120 min after an oral 75 g glucose load were obtained. We measured HbA1c, plasma glucose, total cholesterol, low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C), triglyceride (TG), creatinine, and uric acid using commercial equipment per the standardized methods as previously described<sup>15</sup>. WBC counts were obtained using a hemocytometer (Sysmex XE 2100L, Kobe, Japan). The estimated glomerular filtration rate (eGFR) was calculated according to the Modification of Diet in Renal Disease Study equation.

Body mass index (BMI) was defined as weight (in kilograms) divided by height (in meters) squared.

### Questionnaire

A self-administered questionnaire was used to collect information on treatment for cardiovascular diseases and other diseases including DM, hypertension, dyslipidemia, and high uric acid and social habits such as alcohol drinking and smoking. Alcohol drinking was categorized as drinking ( $n=479$ ), sometimes drinking ( $n=1292$ ), or never drinking ( $n=418$ ). Smoking status was divided into two categories (never smoker ( $n=1580$ ) or former smoker ( $n=217$ )=0, current smoker ( $n=392$ )=1) to stratify the participants into two groups. Data on the average number of cigarettes smoked per day and the number of years of smoking were also collected. The Brinkman index was calculated as the number of cigarettes smoked per day multiplied by the number of years of smoking.

### Definition of Groups

Participants were divided into five age groups: 25-,

**Table 1.** Characteristics of the study participants.

Variables	Overall ( <i>n</i> = 2189)	Non-smokers ( <i>n</i> = 1797)	Smokers ( <i>n</i> = 392)	<i>p</i> -value
Age (years)	35.2 ± 7.1	35.2 ± 7.1	35.4 ± 7.0	0.69
Height (cm)	171.9 ± 5.9	171.8 ± 5.9	172.4 ± 5.9	0.12
Weight (kg)	66.3 ± 9.7	66.0 ± 9.4	68.0 ± 10.6	<0.001
Body mass index (kg/m <sup>2</sup> )	22.4 ± 2.9	22.3 ± 2.8	22.8 ± 3.2	<0.001
Systolic blood pressure (mmHg)	113.7 ± 10.3	113.8 ± 10.4	113.2 ± 10.2	0.32
Diastolic blood pressure (mmHg)	68.3 ± 7.7	68.4 ± 7.7	67.6 ± 7.8	0.06
Heart rate (beats/min)	63.0 ± 8.8	63.3 ± 9.0	61.8 ± 7.7	0.002
baPWV (cm/s)	1219 ± 136	1220 ± 137	1215 ± 134	0.52
Hemoglobin A1C (%)	5.2 ± 0.3	5.2 ± 0.3	5.2 ± 0.3	0.99
Fasting plasma glucose (mg/dl)	88.7 ± 5.5	88.7 ± 5.5	88.8 ± 5.6	0.72
1-hour post-load plasma glucose (mg/dl)	112.0 ± 32.5	111.5 ± 31.8	114.3 ± 35.3	0.13
2-hour post-load plasma glucose (mg/dl)	94.6 ± 18.3	94.6 ± 18.1	94.6 ± 19.5	0.97
Total cholesterol (mg/dl)	192.8 ± 32.6	193.0 ± 32.4	192.3 ± 33.7	0.72
LDL-cholesterol (mg/dl)	111.9 ± 28.8	111.6 ± 28.6	113.6 ± 29.9	0.20
HDL-cholesterol (mg/dl)	57.7 ± 12.8	58.6 ± 12.8	53.5 ± 11.7	<0.001
Triglyceride (mg/dl)	97.8 ± 77.1	92.6 ± 61.6	121.7 ± 123.1	<0.001
estimated GFR (mL/min/1.73 m <sup>2</sup> )	83.4 ± 13.0	82.9 ± 12.8	85.9 ± 13.6	<0.001
Uric acid (mg/dl)	6.0 ± 1.2	6.0 ± 1.2	6.1 ± 1.2	0.57
White blood cell count (× 10 <sup>9</sup> /l)	5.3 ± 1.3	5.1 ± 1.2	5.9 ± 1.5	<0.001
Alcohol drinking (%)	479 (21.8%)	348 (19.4%)	131 (33.4%)	<0.001

All values are expressed as mean ± standard deviation or *n* (%).

Abbreviations: baPWV, brachial-ankle pulse wave velocity; LDL, low density lipoprotein; HDL, high density lipoprotein; GFR, glomerular filtration rate.

30-, 35-, 40-, and 45-year-old participant groups. Overweight was defined by a BMI ≥ 25 (kg/m<sup>2</sup>). Obesity was not defined in this study because there were only 35 participants whose BMI were 30 kg/m<sup>2</sup> or greater. Dyslipidemia was defined by a total cholesterol ≥ 220 mg/dL, LDL-C ≥ 140 mg/dL, HDL-C < 40 mg/dL, and/or TG ≥ 150 mg/dL. Alcohol drinking and smoking status were defined as mentioned above.

### Statistical Analysis

All data were analyzed using STATA 15.1 software (STATA Corp. LLC, College Station, TX, USA). All continuous values are reported as means ± standard deviations, and categorical variables are expressed as percentages. Normally distributed continuous variables were examined using the *t*-test, and non-normally distributed continuous variables were examined using the Mann–Whitney *U* test. All categorical variables were analyzed using Pearson's chi-square test. TG levels and WBC count were log-transformed to improve the normal distribution before the analysis. Correlations between baPWV and the variables were calculated using linear regression analyses. Age, BMI, SBP, DBP, heart rate, fasting plasma glucose, 1h-PG, 2h-PG, LDL-C, HDL-C, log-transformed TG, eGFR, uric acid, log-

transformed WBC, smoking status, and alcoholic drinking were included in the multivariate linear regression analysis model. The number of cigarettes smoked per day or the Brinkman index was added for further analyses. Multicollinearity among explanatory variables was assessed using a variance inflation factor. Product interaction terms were built to test interactions between 1h-PG and other factors including age groups, overweight, dyslipidemia, smoking status, and alcohol drinking in relation to baPWV. An interaction between 1h-PG and smoking status with respect to the log-transformed WBC was also examined. All *p*-values were two-tailed. *P* values less than 0.05 were considered statistically significant.

### Results

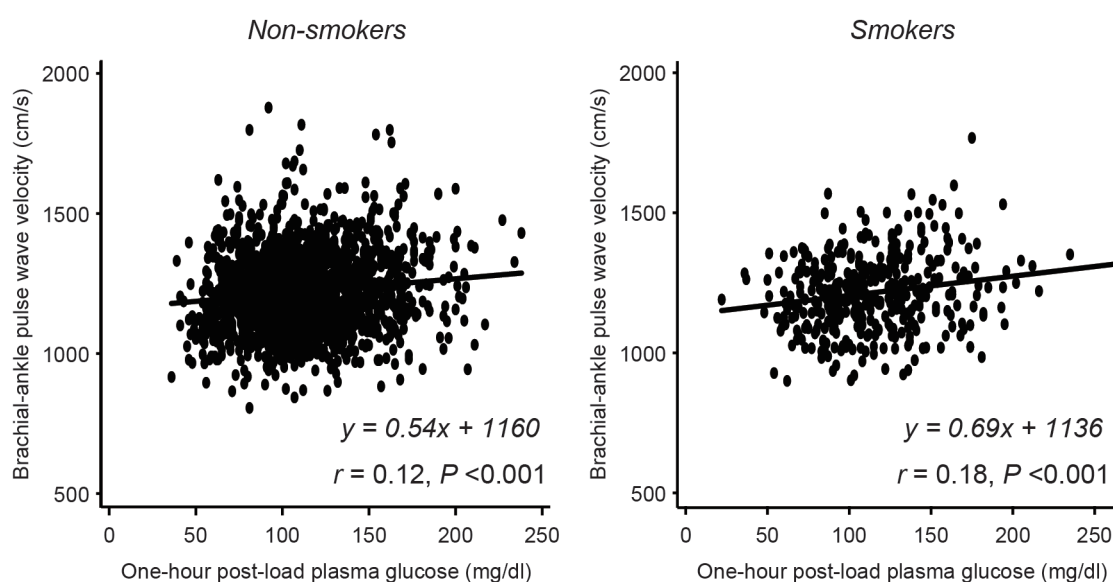
**Table 1** shows the baseline clinical characteristics of the study population. The mean ages of nonsmokers and smokers were 35.2 ± 7.1 and 35.4 ± 7.0 (*p* = 0.69). There were no significant differences in SBP, glucose metabolic profiles, or uric acid. Smoking participants had higher BMIs and TG levels and lower HDL-C levels. As expected, the WBC counts of smokers were higher than those of nonsmokers (5.1 ± 1.3 vs. 5.9 ±

**Table 2.** Univariate linear regression analysis on baPWV as dependent variable in nonsmokers and smokers

Variables	Non-smokers ( <i>n</i> = 1797)		Smokers ( <i>n</i> = 392)	
	<i>r</i>	<i>P</i> value	<i>r</i>	<i>P</i> value
Age (years)	0.28	<0.001	0.26	<0.001
Body mass index (kg/m <sup>2</sup> )	0.08	<0.001	0.09	0.07
Systolic blood pressure (mmHg)	0.39	<0.001	0.36	<0.001
Diastolic blood pressure (mmHg)	0.45	<0.001	0.42	<0.001
Heart rate (beats/min)	0.32	<0.001	0.26	0.004
Fasting plasma glucose (mg/dl)	0.17	<0.001	0.08	0.10
1-hour post-load plasma glucose (mg/dl)	0.12	<0.001	0.18	<0.001
2-hour plasma post-load glucose (mg/dl)	0.10	<0.001	0.12	0.02
LDL-cholesterol (mg/dl)	0.17	<0.001	0.07	0.15
HDL-cholesterol (mg/dl)	-0.02	0.41	-0.08	0.13
Log-transformed TG	0.19	<0.001	0.26	<0.001
estimated GFR (mL/min/1.73 m <sup>2</sup> )	-0.11	<0.001	-0.06	0.25
Uric acid (mg/dl)	0.11	<0.001	0.10	0.0499
Log-transformed WBC	0.09	<0.001	0.15	0.004
Alcohol drinking	0.10	<0.001	0.12	0.02

*r* expresses correlation coefficient.

Abbreviations: baPWV, brachial-ankle pulse wave velocity; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglyceride; GFR, glomerular filtration rate; WBC, white blood cell.



**Fig. 1.** Correlation of 1 h post-load plasma glucose with brachial-ankle pulse wave velocity in nonsmokers and smokers.

1.5,  $p < 0.001$ ). There was no difference in PWV values between nonsmokers and smokers.

#### Interaction between 1h-PG and Smoking in Relation to baPWV and WBC

Among all the participants, a univariate analysis showed that the 1h-PG significantly correlated with

baPWV ( $r = 0.13$ ,  $p < 0.001$ ). Although the correlation between 1h-PG and baPWV was not significant in the multivariate analysis ( $\beta = 0.02$ ,  $p = 0.24$ ), we found a significant interaction between 1h-PG levels and smoking status ( $p$  for interaction = 0.048). There was no significant interaction between 1h-PG levels and age groups ( $p = 0.55$ ), overweight ( $p = 0.66$ ), dyslipidemia

**Table 3.** Multivariate linear regression analysis on baPWV as dependent variable in nonsmokers and smokers

Variables	Non-smokers ( <i>n</i> = 1797)		Smokers ( <i>n</i> = 392)	
	$\beta$	<i>P</i> value	$\beta$	<i>P</i> value
Age (years)	0.18	<0.001	0.17	0.002
Body mass index (kg/m <sup>2</sup> )	-0.13	<0.001	-0.18	0.007
Systolic blood pressure (mmHg)	0.20	<0.001	0.25	<0.001
Diastolic blood pressure (mmHg)	0.20	<0.001	0.19	0.002
Heart rate (beats/min)	0.17	<0.001	0.14	0.005
Fasting plasma glucose (mg/dl)	0.05	0.01	0.004	0.92
1-hour post-load plasma glucose (mg/dl)	0.01	0.79	0.11	0.02
2-hour plasma post-load glucose (mg/dl)	-0.04	0.10	-0.01	0.91
LDL-cholesterol (mg/dl)	0.04	0.11	-0.01	0.89
HDL-cholesterol (mg/dl)	-0.003	0.92	-0.09	0.04
Log-transformed TG	0.07	0.007	0.08	0.25
estimated GFR (mL/min/1.73 m <sup>2</sup> )	-0.03	0.15	-0.03	0.62
Uric acid (mg/dl)	0.04	0.10	-0.01	0.81
Log-transformed WBC	0.04	0.08	0.04	0.39
Alcohol drinking	0.03	0.19	0.07	0.13

$\beta$  expresses standardized beta coefficient.

Abbreviations: baPWV, brachial-ankle pulse wave velocity; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglyceride; GFR, glomerular filtration rate; WBC, white blood cell.

( $p=0.47$ ), or alcohol drinking ( $p=0.11$ ) in relation to baPWV. There was no significant interaction between 1h-PG levels and smoking status in relation to the log transformed WBC ( $p=0.83$ ).

### Correlation between baPWV and 1h-PG in Non-smokers and Smokers

The analyses were conducted in two groups classified according to smoking status (nonsmokers: never or former smokers; smokers: current smokers).

The univariate linear regression analysis revealed that 1h-PG correlated with baPWV in both nonsmokers and smokers (Table 2, Fig. 1), but the correlation coefficient of smokers ( $r=0.18$ ,  $p<0.001$ ) was high compared with that of nonsmokers ( $r=0.12$ ,  $p<0.001$ ).

The multivariate linear regression analysis revealed that 1h-PG significantly correlated with baPWV in smokers ( $\beta=0.11$ ,  $p=0.02$ ), but not in nonsmokers ( $\beta=0.01$ ,  $p=0.79$ ) (Table 3). On the other hand, fasting plasma glucose significantly correlated with baPWV only in nonsmokers ( $\beta=0.05$ ,  $p=0.01$ ).

A sensitivity analysis was performed by dividing nonsmokers into never smokers ( $n=1580$ ) and former smokers ( $n=217$ ) to eliminate the effect of former smokers on the association in nonsmokers. Similar findings were obtained in never smokers: fasting plasma glucose ( $\beta=0.05$ ,  $p=0.02$ ), but not 1h-PG ( $\beta=0.005$ ,  $p=0.84$ ), was significantly associated with baPWV. There was no significant association between baPWV and fasting plasma glucose, 1h-PG, or 2h-PG in former

smokers.

The significant correlation between 1h-PG and baPWV in smokers could be confounded by the extent of smoking because smoking has detrimental effects on glucose metabolism. In fact, 1h-PG was significantly correlated with the number of cigarettes smoked per day ( $r=0.16$ ,  $p<0.001$ ) and the Brinkman index ( $r=0.19$ ,  $p<0.001$ ). Thus, the number of cigarettes smoked per day or the Brinkman index was added into the multivariate linear regression models for smokers. As a result, 1h-PG significantly correlated with baPWV even after adjustment for the number of cigarettes smoked per day ( $\beta=0.096$ ,  $p=0.048$ ) or the Brinkman index ( $\beta=0.097$ ,  $p=0.043$ ).

### Discussion

In the present study, we found a significant interaction between 1h-PG and smoking, but not with other factors in relation to baPWV. By stratifying the participants into non-smoking and smoking groups, the correlations of 1h-PG with baPWV became evident among smokers, but not among nonsmokers. Furthermore, the correlation remained significant even after an adjustment for the extent of smoking, suggesting that 1h-PG correlated with baPWV independent of the dose-dependent effects of smoking on glucose metabolism.

To the best of our knowledge, this is the first study to show that elevated 1h-PG and smoking syn-

ergistically affect arterial stiffness. We previously investigated the associations between 1h-PG and baPWV by stratifying young and middle-aged participants (25 to 55 years) according to their age and sex<sup>15</sup>). Although there was no significant association in women, we found that the association became more evident with aging in men and was significant in men in the 55-year-old age group. This finding may lead to the assumption that the early stages of impaired glucose metabolism do not affect arterial stiffness in young people. However, it is reasonable that the effect of high 1h-PG on vascular damage represents a cumulative effect in young people<sup>19</sup>) even though the effect is not reflected as an increase of arterial stiffness. Thus, the findings of this present study are compelling because the detrimental effects of high 1h-PG on arterial stiffness can be observed even in people younger than 50 years of age if there is a synergistic factor such as smoking. The fact that we only found a significant interaction between 1h-PG and smoking among all the factors including age, overweight, dyslipidemia, and alcohol drinking was also compelling and informative, because smoking is a major but avoidable cause of cardiovascular diseases worldwide<sup>1</sup>).

The underlying mechanisms of the synergistic effect remain unclear. DM and smoking increased arterial stiffness through various common mechanisms<sup>20, 21</sup>). When examining the effect of high 1h-PG, it is important to focus on some of the mechanisms through which arteries stiffen by exposure to rapid increase in plasma glucose, such as oxidative stress, endothelial dysfunction, and subclinical chronic inflammation<sup>19, 22-24</sup>). These detrimental effects were observed even in NGT people<sup>19, 24</sup>) and seem to be related to the plasma glucose level reached<sup>24</sup>). In people with high 1h-PG, inflammatory markers including WBC may be elevated<sup>25, 26</sup>). Thus, we examined WBC count as an index of subclinical chronic inflammation and found that smokers had higher WBC counts than nonsmokers as expected<sup>27</sup>). However, we did not find a significant interaction between 1h-PG and smoking in relation to WBC, suggesting that subclinical chronic inflammation, at least as reflected by WBC, is not a mechanism of the synergistic effect of 1h-PG and smoking on arterial stiffness. Other factors such as oxidative stress and endothelial dysfunction warrant further studies to elucidate the underlying mechanisms of the synergistic effects of 1h-PG and smoking.

Smoking deteriorates glucose metabolism through various mechanisms such as insulin resistance<sup>28</sup>). Thus, smoking increases arterial stiffness<sup>6, 7</sup>) and may increase 1h-PG value simultaneously<sup>29</sup>). Unfortunately, we could not examine insulin resistance, because the health check-up program did not include insulin level measurements.

The 1h-PG values of smokers were slightly higher than those of nonsmokers, although this was not significant ( $p=0.13$ ). The 1h-PG result significantly correlated with the extent of smoking. This is consistent with a finding that insulin resistance develops with increased smoking<sup>30</sup>). In this context, smoking can be a confounder for 1h-PG in relation to baPWV in smokers. Therefore, we performed an additional multivariate analysis and found that the 1h-PG correlated with the baPWV independent of the extent of smoking. Although further analysis that takes insulin levels into account is required, 1h-PG probably would affect the baPWV independent of the detrimental effects of smoking on glucose metabolism in regular smokers.

In contrast with 1h-PG, fasting plasma glucose levels were significantly associated with baPWV in nonsmokers, but not in smokers. A potential explanation for this based on our findings is that fasting plasma glucose levels influence arterial stiffness especially in healthy people, such as the young male nonsmokers in this study. This is consistent with the findings of our previous report, which showed that fasting plasma glucose, but not 1h-PG, significantly correlated with baPWV, but only in young to middle-aged women<sup>15</sup>). Young and middle-aged women are generally at a lower risk of atherosclerosis than men of similar ages, and the smoking rate in the female participants of the study was very low (6.4%). In this context, fasting plasma glucose and 1h-PG probably reflects different aspects of impaired glucose metabolism. Pathophysiological mechanisms behind this, however, remain unclear because insulin levels are required to elucidate these mechanisms and since fasting plasma glucose and 1h-PG levels are largely determined by insulin resistance and insulin secretion abilities. In fact, several studies have revealed that the degree of impairment of glucose metabolism reflected by 1h-PG is not homogeneous when considering insulin profiles<sup>29, 31</sup>). In addition, the clinical significance of this association is limited because the interaction between fasting plasma glucose and smoking in relation to baPWV was not significant (data not shown), and participants whose fasting plasma glucose levels were 100 mg/dL or higher than 100 mg/dL were excluded from this study.

Because arterial stiffness is an independent predictor of cardiovascular diseases and mortality<sup>3, 4</sup>), the synergistic effects of DM and smoking on cardiovascular diseases and mortality are also expected to be observed. However, no study has shown a significant interaction between smoking and DM on the risks of cardiovascular events and mortality<sup>30, 32</sup>). A possible explanation is that the findings of this study are applicable only to people that are in the early stages of arterial stiffening, because this study included young and healthy people

with relatively low baPWV values ( $1219 \pm 136$  cm/s) compared with the cutoff value (1591 cm/s) for predicting future cardiovascular disease risk, as examined by an individual participant data meta-analysis<sup>4)</sup>.

We found a significant interaction between 1h-PG and age in our previous study<sup>15)</sup>, but the interaction was not significant in this present study. The exclusion of 50- to 55-year-old people could be a reason for this inconsistency because the association between 1h-PG and baPWV was more evident in the participants of the former study than in the participants of this study.

Our study has some potential limitations. First, our study is cross-sectional. Therefore, a causal relationship between 1h-PG and smoking and arterial stiffness could not be determined. Second, as mentioned above, insulin levels were not accounted for in this study because of the lack of available data. We have carefully adjusted for the potential confounders, but we could not include the insulin-related indices. Thus, it is possible that there is residual confounding of this unmeasured variable. Third, the large samples size comprising both nonsmoking and smoking relatively young men was a major strength of this study, but the exclusion of women was another limitation. Finally, the study was performed in a Japanese population of participants. Further studies of other ethnic groups are warranted to generalize our findings.

In conclusion, we found a synergistic effect between 1h-PG and smoking on baPWV in male NGT participants younger than 50 years of age. Although the risk of cardiovascular diseases is considered to be very low in these people, the harmful effects of high 1h-PG on arterial stiffness can be enhanced by smoking and become evident. In addition, because smoking is a major but avoidable cause of cardiovascular diseases, the findings of this study can be beneficial to young people by providing an opportunity for the recognition of the synergistic and detrimental effect of smoking on vascular damage.

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### Conflicts of Interest

Yoshio Kobayashi received lecture fees from Astellas AMGEN Biopharma (California, United States), Astra-Zeneca (Cambridge, England), Sanofi (Paris, France), Daiichi-Sankyo (Tokyo, Japan), Bristol-Myers Squibb (New York, United States), and Boehringer Ingelheim

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