Augmentation index is a better marker for cardiovascular risk in young Malaysian males

A comparison of involvement of pulse wave velocity, augmentation index, and C-reactive protein

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ABSTRACT

Objectives: To determine the association between carotid femoral pulse wave velocity (PWVCF) and augmentation index (AI) with future cardiovascular disease (CVD) risk, and to assess whether high sensitivity C-reactive protein (hs-CRP) is an important mediator towards these vascular changes, among young men.

Methods: This cross-sectional study was conducted at Universiti Kebangsaan Malaysia Medical Center, Kuala Lumpur, Malaysia from July 2011 to December 2012. Two hundred and eleven young men were recruited. The PWVCF and AI were measured using Vicorder. High sensitivity C-reactive protein was measured by using immunological methods. The future CVD risk was assessed by Framingham risk score (FRS) and age adjusted FRS (A-FRS). Data for analysis was conducted using the Statistical Package of Social Sciences Version 15 (SPSS Inc., Chicago, IL, USA).

Results: The mean age of the subjects was 27.09 (95% confidence intervals [CI] 26.39-27.79) years old. Those with ≥2 risk factors had significantly higher AI [10.09 (95% CI: 9.06-11.12) versus 6.56 (95% CI: 5.54-7.57) (P=0.001), but not PWVCF 7.45 (95% CI: 7.29-7.61) m/s versus 7.29 (95% CI: 7.06-7.51) m/s, (P=0.90) when compared to the healthy subjects. High sensitivity C-reactive protein was not an independent determinant for PWVCF and AI. Only AI was significantly associated with FRS and A-FRS (P=0.0001).

Conclusions: To assess the impact of risk factors on vascular damage and for future assessment of CVD risk among the young men, AI may be a better marker than PWVCF. The increase in AI among these subjects was not related to hs-CRP.
Pulse wave velocity (PWV) and augmentation index (AI), as tools for the assessment of vascular damage are widely used. Both parameters are easily determined, reproducible, noninvasive and safe, and have been shown to predict cardiovascular disease (CVD) morbidity and mortality. Because of these features, they are suitable for general health screening procedures, especially among young subjects. By exploring the impact of cardiovascular (CV) risk factors on vascular function, an early marker of vascular damage can be identified so that preventive measures can be taken at an early stage. The measurement of aortic PWV and AI are based on the analysis of aortic pressure wave. The aortic pulse pressure (PP) or central PP (cPP) constitutes a forward pressure wave and reflected wave. Forward pressure wave is represented by the height of the first systolic pressure (P1) and is related to ventricular ejection and stiffness of the aorta. One of the measures of aortic stiffness (AS) is carotid femoral PWV (PWVCF), which reflects the velocity of the forward wave traveling along the aorta to the femoral artery. Pulse wave velocity increases as the stiffness of the aorta increases. As the forward wave travels, part of it is reflected back to the aorta when they hit the peripheral artery branches, and augments the pressure in the aorta (augmentation pressure [AP]). Augmentation pressure is defined as the difference of pressure between the second systolic peak and first systolic peak. Augmentation index (AI) is a term representing the wave reflection and is calculated as AP over central pulse pressure (cPP) x 100. Recently, AP was found to be the main component of increased aortic PP due to aging among young healthy females instead of P1. The value of AI would be increased if PWV increased. Thus, a consensus suggested that AI is a surrogate measure of AS. However, AI is also influenced by other physiological factors, such as peripheral vasodilation, height and heart rate (HR) of the subjects, which may mask its association with PWV. Because of these factors, the impact of CVD risk factors on both PWV and AI in the same subjects may also be different. Several studies observed that both aortic PWV and AI increased in those with risk factors, while other studies observed that only aortic PWV was increased and not AI. These discrepancies may be due to different factors affecting both parameters as mentioned above.

In addition, the effects of risk factors on PWV and AI may also be influenced by age. Some studies suggested that AI is a sensitive marker of vascular damage among the young subjects. Another review also found aortic PWV was strongly associated with hypertension and has weak associations with other major CVD risk factors, which may not be sensitive as an early marker of vascular damage among non-hypertensive young subjects. However, all these studies did not extend their findings by comparing the association between PWV and AI with future coronary heart disease (CHD) risk, such as Framingham Risk Score (FRS). Although many cohort/prospective studies have been conducted, and it was observed that these markers are predictor of future CVD morbidity and mortality among those with various diseases, these observations were obtained from middle aged and older subjects. Due to the length of time taken to see mortality rate in the young, such predictions are lacking. As an alternative, CHD risk score to determine future risk of CHD such as FRS can be used. Few studies have addressed this issue, and were mainly on middle age and older subjects which revealed that PWV had stronger relation with CV risk compared to AI. In contrast, another study that involved 144 subjects without atherosclerotic disease (mean age: 46±14 years old) found that AI was strongly correlated with European Society of Cardiology (ESC) risk score. Studies among the young subjects are scarce. In one study, it was found that only PWV was significantly associated with FRS score and not AI in men.

The development of atherosclerosis involves inflammatory mediators in every stage. Among the inflammatory mediators involved are interleukin (IL)-1β, IL-18, IL-6, tumor necrosis factor (TNF) α and β, monocyte chemo-attractant protein-1 (MCP-1) and monocyte colony stimulating factor (MCSF), which are secreted by the smooth muscle cells and monocytes. C-reactive protein (CRP) is systemic inflammatory marker which is secreted by the liver after activation by IL-6. Inflammatory mediators especially CRP are also thought to play an important role in increasing arterial stiffness by several mechanisms such as 1) inducing the release of matrix metalloproteinases (MMP) from the leukocytes, which cause elastin degradation, 2) cause endothelium dysfunction by inactivation of nitric oxide (NO) and decreased endothelium NO synthase (eNOS) expression, 3) increased smooth muscle proliferation, and 4) causing vascular calcification. Inflammation, while causing an increase in PWV by several mechanisms, may reduce AI by peripheral vasodilation. However, studies among the middle age and older subjects found that increased inflammation

Disclosure. This study was funded by the Universiti Kebangsaan Malaysia, Kuala Lumpur, Malaysia. (FF-262-2011). Authors have no conflict of interests, and the work was not supported or funded by any drug company.
was associated with increased AI.\textsuperscript{20,21} Studies among the young are lacking and need to be investigated. Such research is valuable in providing basic information in the pathophysiology of vascular damage. The current study was conducted to determine the association between aortic PWV\textsubscript{CF} and AI with future CVD risk. Secondly, to determine whether CRP is an important mediator of any changes on aortic PWV\textsubscript{CF} and AI due to CV risk factors. In this study, young males are the main focus since this group is more vulnerable to CVD compared with females.\textsuperscript{22}

**Methods.** A search of previous publications was carried out by internet websites such as Google Scholar, Ovid, and PubMed. This cross sectional study was approved by the Universiti Kebangsaan Malaysia Medical Center Ethics Committee (FF-262-2011). The subject recruitment commenced from July 2011 to December 2012. Subjects were recruited from areas around Klang Valley, Klang Valley is an area which includes the capital city Kuala Lumpur and its suburbs. The main study was conducted at Universiti Kebangsaan Malaysia Medical Center, Kuala Lumpur, Malaysia. Subjects were also recruited by screening programs held in various places in Klang Valley. It used a purposive sampling method, with careful subject selection. All the subjects gave written consent and this study followed the principles of the Helsinki Declaration.

The inclusion criteria were young men aged 20-39 years old, healthy, or may have any cardiovascular risk factors such as hypertension, dyslipidemia, abdominal obesity, smoking, and family history (FH) of CVD. Exclusion criteria were those with diabetes mellitus and other chronic disease such as CVD, peripheral vascular disease, lung disease, liver disease, and inflammatory disease. Diabetes mellitus was excluded since this disease is equivalent to coronary artery disease (CAD), and subjects may have advanced vascular damage compared to other CV risk factors.\textsuperscript{12} Criteria for young Malaysian males for various CV risk factors was observed as per reference given with each of the following: 1) Hypertension: systolic blood pressure ≥140 and/or diastolic ≥90 or on antihypertensive medication.\textsuperscript{23} 2) Diabetes mellitus: fasting plasma glucose ≥7mmol/L.\textsuperscript{24} 3) Smokers: a habit of daily smoking continued at the time of recruitment for study.\textsuperscript{23} 4) Abdominal obesity: waist circumference >90 cm.\textsuperscript{26} 5) Family history (FH) of premature CAD: when parents had CAD at <55 (father) or <65 (mother) age.\textsuperscript{12,6} 6) Dyslipidemia: when TC >6.2mmol/L, TG >1.7mmol/L, LDL >4.2mmol/L, or HDL <1.04mmol/L.\textsuperscript{27}

In this study, only one subject was on medication, which was for dyslipidemia. Approximately 70\% of the subjects were Malay and the rest were Malaysian Chinese. They were grouped into 1) those with 0 risk factor, 2) those with one risk factor, and 3) those with ≥2 risks. The minimum sample size was 50 subjects per group and was calculated manually by using a published formula.\textsuperscript{28} It was estimated on the bases of 80\% power and 95\% confidence interval (CI), referring the values of PWV and AI from a previous study.\textsuperscript{6} At the end of the study, a total of 211 young men were recruited.

**Measurement of body anthropometry.** Height was measured by a wall-mounted stadiometer (SECA, Hamburg, Germany) and weight was measured by using a digital scale (SECA, Hamburg, Germany). Body mass index was then calculated as weight (kg)/height (m\(^2\)). Waist circumference was measured by a measuring tape on the horizontal plane, midway between the anterior superior iliac spine and lower rib after normal expiration.\textsuperscript{27}

**Measurement of carotid femoral pulse wave velocity and AI.** The measurements of both parameters were carried out by using a Vicorder (SMT Medical, Wuerzburg, Germany) in supine position. For PWV\textsubscript{CF} measurement, a neck cuff that contained pressure sensor for the carotid artery was placed around the neck. Another cuff was placed around the subjects’ right thigh which sensed pressure from the femoral artery. A distance (D) between sternal notch and mid thigh cuff was then measured (in meter [m]) by using a measuring tape. After lying for 10 minutes, the measurement was initiated by inflating the neck cuff and the thigh cuff to 65 mm Hg simultaneously, to get the corresponding oscillometric signals. The signals were analyzed to get the delays between the 2 recorded pulses (transit time [TT]). Pulse wave velocity of the carotid femoral was then calculated as D over TT (m/s).

For the measurement of AI, a brachial cuff was placed on the right arm, which detected the brachial pressure waveform when it was inflated. Then, by using a brachial-to-aortic generalized transfer function, the central aortic pressure waveform was derived. Augmentation index was calculated as [(second systolic peak-first systolic peak)/pulse pressure x 100].\textsuperscript{1} The brachial BP was also obtained by using this device. For all the parameters, every subject underwent one measurement.

**Framingham risk score.** To estimate the FRS, the formula\textsuperscript{29} was adopted and the risk factors that were taken into account were age, blood pressure, total cholesterol, HDL cholesterol, diabetes status, and smoking. Each risk factor had a score point based on
the severity. In the current study, FRS was calculated by using a FRS calculator, and the total points of FRS were used in the results and analysis. Since age is an important component of FRS, this will give a bias in our subjects because of the wide range of age, thus, another parameter was added. The age adjusted FRS (A-FRS) was derived by subtracting the total FRS points with the age point of each individual.\(^{31}\)

**Measurement of blood parameters.** Blood was withdrawn from the antecubital vein after fasting for a minimum of 8 hours. Blood samples were then sent to Gribbles pathology laboratory (Petaling Jaya, Selangor, Malaysia) for further analysis of lipid profiles, hs-CRP, and glucose. This laboratory obtained International Organization of Standardization (ISO: MS ISO 15189) in compliance with the standard quality. The serum TG, HDL cholesterol, and TC were measured using enzymatic methods (Advia 2400 Chemistry Analyzer, Siemens, Tokyo, Japan). The hs-CRP level was measured by immunological methods (Advia 2400 Chemistry Analyzer, Siemens, Tokyo, Japan). The blood glucose was measured by enzymatic method using hexokinase and glucose-6-phosphate dehydrogenase enzymes (Advia 2400 Chemistry Analyzer, Siemens, Tokyo, Japan). For lipids profile, the inter-assay coefficient of variant (CV) ranged from 1.4-3.5%. The inter-assay CV for hs-CRP ranged between 2-2.4%, and CV for glucose ranged from 1.6-1.7%.

**Statistical analysis.** Visual inspection of the histogram (plotted as the distribution frequencies) and acceptable level of skewness (-1 to 1) and kurtosis (-1 to 1) were used to determine the normality of the data. All the data were normally distributed except for hs-CRP, which was skewed. The values of hs-CRP were logarithmically transformed to improve the skewness and were used in data analysis. All the data were in mean (95% CI) except for hs-CRP, which was in median [inter quartile range (IQR)]. The differences in biophysical profiles between groups were analyzed by analysis of variance (ANOVA). The levels of AI and PWV\(_{\text{CF}}\) between groups were compared by general linear model (GLM) univariate to adjust for confounders; age, HR, height, and race. Post hoc analysis was conducted for AI using GLM univariate with Bonferroni adjustment to determine the difference between each group.\(^{32}\) The correlations between PWV, AI, and hs-CRP were carried out by Pearson correlation (\(r\)). The independent determinants for AI and PWV\(_{\text{CF}}\) were determined by multiple linear regression and adjusted coefficient of determinants (R\(^2\)) was used to represent the amount of variance that was contributed by the independent parameters. To determine the association between increased PWV\(_{\text{CF}}\) and AI with FRS and A-FRS, PWV\(_{\text{CF}}\) and AI were divided into lowest, second, and highest tertile, and the FRS and A-FRS points in each tertile were compared by analysis of variance. To determine the strength of association between PWV\(_{\text{CF}}\) and AI with FRS and A-FRS score, linear regression was carried out. The standardized regression coefficient (\(\beta\)) was used to explain the change in FRS and A-FRS (points) per one unit increase in PWV\(_{\text{CF}}\) (in m/s) and AI (in %). The significant results were accepted as \(p<0.01\) for post hoc analysis and \(p<0.05\) for others. All the data were analyzed using the Statistical Package for Social Sciences Version 15 (SPSS Inc., Chicago, IL, USA).

**Results.** The subjects’ characteristics for the whole and each group are summarized in Table 1. They were young males (n=211), with mean BP, WC, BMI, lipid profile and blood sugar within normal range. The hs-CRP level was considered to be in the lower range value, which is ≤1mg/L.\(^{33}\) The prevalence of hypertension was 10.7%, abdominal obesity 33%, dyslipidemia 46.4%, smoker 33.3%, and FH of CAD 4.9%. None of them had diabetes mellitus or prediabetes (6.1 mmol/L, <FBS <7mmol/L). There were significant increasing trends in terms of their age, BP, WC, BMI, cholesterol level, hs-CRP level, FRS and adjusted FRS from 0 risk to many risks.

Table 2 depicts the vascular properties in each group. After adjustment for the age, race, heart rate and height, the value of PWV\(_{\text{CF}}\) was insignificantly different between groups (\(p=0.90\)), while for AI, there were significant increasing trends from 0, one and ≥2 risk factors (\(p=0.001\)). For AI, post hoc analysis revealed that significant difference existed between groups with ≥2 risk factors versus groups with 0 risk factors (\(p=0.004\)) after adjustment for confounders. An insignificant difference was obtained between 0 risk factor versus 1 risk factor (\(p=0.74\)). Augmentation pressure was significantly different between groups (\(p=0.02\)), while no difference was observed for P1 (\(p=0.07\)). Correlation analysis revealed that hs-CRP correlated significantly with AI (\(r=0.2, p=0.005\)), and no correlation exists between hs-CRP and PWV\(_{\text{CF}}\) (\(p=0.44\)). Multiple regression analysis revealed that MAP and TG were independent determinants for PWV\(_{\text{CF}}\) (adjusted R\(^2\)=0.11) while AI was independently determined by age, WC, HR, MAP, smoker and blood sugar (adjusted R\(^2\)=0.40). High sensitivity C-reactive protein failed to be the independent determinant of AI. The significant correlation obtained between hs-CRP and AI by Pearson correlation analysis as mentioned above may be confounded by other factors such as age.
Figures 1 and 2 depicts the level of FRS and A-FRS points as the level of AI and PWV\textsubscript{CF} increased. The FRS and A-FRS points were only significantly increased when the level of AI increased ($p=0.0001$) for both and not PWV\textsubscript{CF}. In linear regression, the association between AI and FRS ($\beta=0.47$) and A-FRS ($\beta=0.43$) were significant ($p=0.001$ for both). No significant association was observed between PWV\textsubscript{CF} with FRS ($\beta=0.11$, $p=0.13$) and PWV\textsubscript{CF} with A-FRS ($\beta=0.09$, $p=0.19$).

**Discussion.** The current study showed 3 main findings. First, AI is a more sensitive marker of vascular damage compared to PWV\textsubscript{CF} among the young subjects with risk factors; however, these can be detected only in those with 2 or more risks. Augmentation index also has significant association with future cardiovascular risk and lastly, the increase in AI is not mediated by hs-CRP.

Table 1 - Characteristics of the young males (n=211) with mean blood pressure, WC, BMI, lipid profile and blood sugar.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Total (N=211)</th>
<th>0 risk (n=61)</th>
<th>One risk (n=56)</th>
<th>≥2 risk (n=94)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean (95% CI)</td>
<td>Mean (95% CI)</td>
<td>Mean (95% CI)</td>
<td>Mean (95% CI)</td>
<td>Mean (95% CI)</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>27.09 (26.39-27.79)</td>
<td>24.80 (23.57-26.03)</td>
<td>26.90 (25.46-28.34)</td>
<td>28.72 (27.76-29.68)</td>
<td>0.0001</td>
</tr>
<tr>
<td>SBP (mm Hg)</td>
<td>126.86 (125.52-128.21)</td>
<td>123.75 (121.66-125.84)</td>
<td>126.42 (123.83-129.02)</td>
<td>129.19 (127.02-131.35)</td>
<td>0.003</td>
</tr>
<tr>
<td>DBP (mm Hg)</td>
<td>72.88 (71.88-73.89)</td>
<td>70.67 (68.92-72.42)</td>
<td>72.24 (70.35-74.12)</td>
<td>74.73 (73.17-76.29)</td>
<td>0.003</td>
</tr>
<tr>
<td>MAP (mm Hg)</td>
<td>94.25 (93.14-95.36)</td>
<td>91.24 (89.44-93.04)</td>
<td>93.15 (91.08-95.22)</td>
<td>96.91 (95.18-98.63)</td>
<td>0.0001</td>
</tr>
<tr>
<td>HR (bpm)</td>
<td>65.76 (64.38-67.13)</td>
<td>65.37 (62.85-67.88)</td>
<td>63.58 (60.67-66.48)</td>
<td>67.37 (65.39-69.34)</td>
<td>0.08</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>86.91 (85.26-88.56)</td>
<td>78.75 (77.12-80.38)</td>
<td>83.74 (81.32-86.16)</td>
<td>94.06 (91.46-96.66)</td>
<td>0.0001</td>
</tr>
<tr>
<td>BMI (kg/m(^2))</td>
<td>24.57 (23.90-25.23)</td>
<td>21.29 (20.64-21.93)</td>
<td>23.27 (22.35-24.19)</td>
<td>27.49 (26.41-28.57)</td>
<td>0.0001</td>
</tr>
<tr>
<td>TC (mmol/L)</td>
<td>5.10 (4.98-5.21)</td>
<td>4.78 (4.61-4.96)</td>
<td>4.95 (4.76-5.14)</td>
<td>5.38 (5.19-5.57)</td>
<td>0.0001</td>
</tr>
<tr>
<td>TG (mg/dL)</td>
<td>1.21 (1.11-1.30)</td>
<td>1.04 (0.76-0.92)</td>
<td>1.21 (0.93-1.19)</td>
<td>1.53 (1.36-1.69)</td>
<td>0.0001</td>
</tr>
<tr>
<td>HDL (mmol/L)</td>
<td>1.18 (1.14-1.22)</td>
<td>1.36 (1.30-1.41)</td>
<td>1.20 (1.13-1.27)</td>
<td>1.05 (1.00-1.11)</td>
<td>0.0001</td>
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<tr>
<td>LDL (mmol/L)</td>
<td>3.36 (3.26-3.46)</td>
<td>3.04 (2.89-3.19)</td>
<td>3.26 (3.09-3.44)</td>
<td>3.62 (3.45-3.78)</td>
<td>0.0001</td>
</tr>
<tr>
<td>FBS (mg/L)</td>
<td>4.66 (4.61-4.72)</td>
<td>4.69 (4.58-4.81)</td>
<td>4.65 (4.54-4.76)</td>
<td>4.65 (4.58-4.72)</td>
<td>0.76</td>
</tr>
<tr>
<td>Hs-CRP (mg/L)</td>
<td>0.60 (2.12)</td>
<td>0.20 (1.08)</td>
<td>0.40 (1.38)</td>
<td>1.30 (2.98)</td>
<td>0.0001</td>
</tr>
<tr>
<td>FRS (points)</td>
<td>0.65 (-0.07-1.37)</td>
<td>-3.97 (-4.79-3.15)</td>
<td>0.76 (-0.27-1.79)</td>
<td>3.60 (2.55-4.65)</td>
<td>0.0001</td>
</tr>
<tr>
<td>A-FRS (points)</td>
<td>9.11 (8.42-9.80)</td>
<td>4.75 (4.40-5.51)</td>
<td>9.07 (8.08-10.06)</td>
<td>11.91 (10.89-12.93)</td>
<td>0.0001</td>
</tr>
</tbody>
</table>

Table 2 - Vascular properties between groups.

<table>
<thead>
<tr>
<th>Values</th>
<th>Total (N=211)</th>
<th>0 risk (n=61)</th>
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<td>Mean (95% CI)</td>
<td></td>
</tr>
<tr>
<td>PWV\textsubscript{CF} (m/s)</td>
<td>7.38 (7.27-7.49)</td>
<td>7.29 (7.06-7.51)</td>
<td>7.36 (7.17-7.56)</td>
<td>7.45 (7.29-7.61)</td>
<td>0.44*</td>
</tr>
<tr>
<td>AI (%)</td>
<td>8.33 (7.66-9.00)</td>
<td>6.56 (5.54-7.57)</td>
<td>7.39 (6.09-8.69)</td>
<td>10.09 (9.06-11.12)</td>
<td>0.001</td>
</tr>
<tr>
<td>AP (mm Hg)</td>
<td>4.07 (3.71-4.43)</td>
<td>3.07 (2.45-3.70)</td>
<td>3.77 (2.98-4.55)</td>
<td>4.80 (4.30-5.31)</td>
<td>0.0001</td>
</tr>
<tr>
<td>P1 (mm Hg)</td>
<td>40.29 (39.45-41.12)</td>
<td>39.80 (38.21-41.39)</td>
<td>39.79 (38.31-41.27)</td>
<td>40.83 (39.52-42.15)</td>
<td>0.48*</td>
</tr>
</tbody>
</table>

*unadjusted, †after adjustment for the age, ††after adjustment for age and race, *after adjustment for the age, race, and heart rate, \*after adjustment for age, race, heart rate, and height, \*p=0.74 versus 0 risk factor, \*p=0.004 versus 0 risk factor, PWV\textsubscript{CF} - carotid femoral pulse wave velocity, AI - augmentation index, AP - augmentation pressure, P1 - the height of the first systolic shoulder (forward wave pressure).
the young using PWV and AI is gaining popularity for screening purposes since the measurements are noninvasive, reproducible, and reliable. Thus, it is important to recognize the pattern of vascular changes among these populations. The current study, which mostly involved non-hypertensive young male, suggests that AI is a better marker of vascular damage due to risk factors of CVD compared to aortic PWV. The increase in AI among the young subjects with risk factors were in line with a previous study. The lack of increased aortic stiffness among the young male subjects with risk factors was also in line with a previous study. This supports studies that suggest PWV is less sensitive compared to AI in examining the impact of risk factors on the vascular system, especially in a non-hypertensive young population. Based on 998 healthy subjects, McEniery et al found that the changes of PWV following an increasing age is more marked among the older subjects, while the changes in AI (4001 healthy subjects) was more pronounced among the younger subjects. They also found that CVD risk factors have greater impact on PWV in the older subjects, while in the young subjects, risk factors have greater impact on AI. In addition, recent systematic review highlighted that high BP is the only risk factor recognized to be strongly correlated with PWV. The higher pressure places more load on the arterial wall and the tension is transferred from elastin to collagen, which cause overall stiffening of the wall (functional stiffness). Chronic elevation of BP may also cause structural damage to the wall, which includes elastin breakdown and increased collagen deposition. In contrast, less association between PWV and other risk factors of atherosclerosis among the young were observed and may be due to the fact that PWV was found to be associated with atherosclerosis when only advanced plaque is present. In the young subjects with risk factors, the development of advanced plaque (example, fibrous deposition and calcification) may take a long time to develop, and measurement of PWV at early stage of risk factors may not identify any abnormality. However, the measurement of PWV cannot be left out among the young with risk factors since the finding of an increased level may signify that the subject may have advanced atherosclerotic plaque. For AI, post hoc analysis revealed that a significant difference was obtained only between 0 and ≥2 risk factors. No difference was observed between 0 and one risk factor. This suggests that AI may not be sensitive enough to detect vascular damage in young subjects with one risk factor. It is expected that those with one risk factor might have minimal changes of vascular damage. This is also in line with the current clinical consensus, which stated that those with 0 and one risk factors are at low risk, and have 10 year risk of CHD that rarely reach levels, which need intensive intervention.

With regards to the aortic pressure waveform, the current study observed that the increase in cPP in the risk factor group was attributed by increase in AP and not P1. Since P1 is associated with aortic stiffness, no difference in P1 further supports the lack of increased aortic stiffness in those with risk factors. It also has another important implication. Studies found that AP can be reduced, hence reducing the cPP by peripheral vasodilators, such as nitroglycerine. Since cPP was
found to be a strong predictor of CVD mortality,\textsuperscript{57} therefore, this method can be used as the target of drug therapy.

Pulse wave velocity, augmentation index, and inflammation. The increased in AI can be attributed to factors such as endothelial dysfunction\textsuperscript{48} and may also be related to the activation of the renin angiotensin system (RAS).\textsuperscript{39} Several other studies also suggested that inflammation may play a role in increased AI.\textsuperscript{21,40} A study carried out among middle aged and older subjects with chronic inflammation (vasculitis) also observed similar findings.\textsuperscript{20} Inflammatory markers, particularly CRP, might cause endothelial dysfunction,\textsuperscript{41} which may increase peripheral vascular resistance and, hence, wave reflection. The current study found that hs-CRP was not an independent determinant of AI, which suggests that no relationship exists between AI and inflammation. Previous studies in young male subjects also observed similar findings.\textsuperscript{35} This was also supported by other different inflammatory models, which showed that acute inflammation, which was induced by giving endotoxin or vaccination, caused either reduction in the level of AI\textsuperscript{19} or gave no effect at all.\textsuperscript{42} The lack of association between AI and hs-CRP in the current study may be due to the low level of hs-CRP in our subjects (<1 mg/L). Another possibility is the effect of age. It was clear that the significant positive associations that were obtained between AI and inflammation were mostly involved middle and older age subjects. Only a few studies among the middle aged and old subjects found that AI was not associated with inflammation.\textsuperscript{7,43} Due to the aging process, the defensive mechanisms significantly deteriorate, a process which is known as immunosenescence.\textsuperscript{44} Aging is also associated with activation of innate immunity, which leads to a pro-inflammatory condition (inflamm-aging).\textsuperscript{44} These indicate that younger subjects have a stronger immune system compared to the older. Thus, their level of inflammation may be much lower and may not be an important cause of any vascular changes, at least at the early stage of atherosclerosis. However, the influence of aging on the association between hs-CRP and AI should be investigated further. The lack of association between AI and hs-CRP also suggests that the increased AI in our subjects was due to other factors as mentioned above. These factors may act by increasing the peripheral vascular resistance, which increase the magnitude of wave reflection, an important component of AI.

Inflammation was known to increase aortic stiffness by triggering a series of events that cause an increase in collagen, fibrous tissue, and reduces elastin.\textsuperscript{45} Studies among the middle aged and older subjects observed positive significant association between inflammation and PWV.\textsuperscript{35,46} However, the current study failed to show such association and was in agreement with previous study in young subjects.\textsuperscript{46} This can be due to the level of hs-CRP and the aortic stiffness itself, which were not high. Another reason is that any structural changes that occur due to the inflammatory process may take a long time to develop and at an early stage, may show a non-significant association. Although one study involving young subjects observed significant association, this association was obtained by univariate analysis, which can be confounded by other factors such as age and blood pressure.\textsuperscript{47}

In the Malaysian population, our previous work observed that Malays had higher AI compared with the Malaysian Chinese, and we proposed to include the race as the confounding factor when statistical analysis was carried out.\textsuperscript{7} However in the current study, the race factor failed to be the independent determinant of AI. This may be due to the low numbers of Malaysian Chinese involved (30%) compared with previous work (72%). Nevertheless, we still included the race as the confounder when statistical analysis was carried out to reduce any bias.

Pulse wave velocity, augmentation index, and future cardiovascular disease risk. The concept of age adjusted FRS was carried out in order to eliminate the influence of age, which has strong association with AI.\textsuperscript{31} Our suggestion to use AI as an early biomarker of CVD among younger subjects is strengthened with the finding of its strong positive association with FRS, which remained significant when analysis was carried out with age adjusted FRS. Our finding is in contrast with another study among 224 young men with CV risk factors, which found that PWV had significant positive association with FRS but not AI.\textsuperscript{16} In their study, the FRS was not adjusted for age as the age range was small (27-30 years old, mean age 28.2±1.0 years old). Although their subjects’ age was nearly similar to our subjects’ mean age (27.09 [95% CI: 26.39-27.79] years old), their subjects’ mean SBP was around 132.2±12.4 mm Hg, which was higher compared with our subjects (126.86 [95% CI: 125.52-128.21] mm Hg). As BP is an important determinant of PWV, the difference in BP between our subjects and theirs’ may be the cause of the discrepancy. The level of AI in our study also was higher (8.33 [95% CI: 7.66-9.00]) compared with theirs (3.17±11.1%), and this can be due to the presence of other risk factors including ethnicity.

In Malaysia, studies on PWV\textsubscript{CF} and AI are limited. Since their values are partly affected by ethnicity, the current study provides reference values of PWV\textsubscript{CF} and
AI among younger males in Malaysia, and will benefit the local researchers. The current study also explored the pathophysiology of vascular damage among younger males. For future research, the different impact of CV risk factors on PWV\textsubscript{CF} and AI should be investigated among the young females, which have different characteristic and hormonal status.

**Study limitations.** We did not venture to measure other biomarkers of inflammation such as IL-6 and TNF-alpha, which may give more comprehensive information on the relationship between inflammation, AI, and PWV. In this study, we did not have reproducibility data for PWV\textsubscript{CF} and AI. The findings were also confined to young males, and may not be applicable to females or older aged groups.

In conclusion, to assess the impact of risk factors on vascular damage and for future assessment of CVD risk among young men, AI may be a better marker than PWV\textsubscript{CF}. The increase in AI among these subjects was not related to hs-CRP.

**Acknowledgment.** The authors would like to express their sincere appreciation to Musmarlina Omar, Fadzilah Suratman, Norizam Salam, Azmir Ahmad, Firdaus Azmi, and Nurlahilla for technical support.

**References**


