Heritable factors play a key role in the development of coronary heart disease. Early unbiased genome-wide approaches have led to the identification of more than a dozen novel risk loci for myocardial infarction (MI) in predominantly Caucasian cohorts. Many loci have replicated in large cohorts, yet of these only four appear to have a mechanistically relevant basis through lipid metabolism pathways (1p13, 1p32, 19p13, 6q26). The remaining loci span genomic regions that as yet have no clearly defined mechanism to explain MI risk.

Valuable insights into the functions of these variants may be gleaned by studying their effects on related or intermediate phenotypes which may be causal in the pathway to developing MI. This is best illustrated by the initial association of 1p13 with MI and then with the intermediate phenotype of lipid levels, with subsequent functional studies identifying its role in cellular cholesterol transport through the sortillin protein. Another intermediate phenotype of interest in vascular disease is abnormal pressure wave reflection within the arterial tree. Abnormalities in these arterial tree properties occur due to complex composite changes in parameters such as arterial stiffness and vasomotor tone, ultimately leading to increased left ventricular after-load and cardiovascular events such as myocardial ischemia, MI, and stroke.

We therefore investigated the association of variants at eight MI risk loci, whose mechanisms of action are currently unknown, with noninvasively derived pulse wave analysis (PWA) indexes.
including the magnitude and timing of reflected arterial pressure waveforms in two separate Caucasian populations. The first consisting of very healthy subjects, free of confounding risk factors to enable an assessment of direct genetic risk and the second, an unselected community-based population that allowed determination of genetic effects in the context of common risk factors. We hypothesized that some or all of these markers would demonstrate association with indexes of adverse arterial wave reflection.

**METHODS**

**Subjects.** Two populations were studied (Table 1). Group 1 was derived from a healthy sample of subjects free of cardiovascular risk factors enrolled in the Predictive Medicine (PreMed) study.16 This group was chosen to minimize confounding from coronary artery disease, medication usage, and other risk factors. Healthy nonsmoking, normal weight (body mass index (BMI) <26 kg/m²) volunteers aged 20–70 years were recruited after careful screening for absence of hypertension (systolic blood pressure (SBP) <130 or diastolic BP (DBP) <90 mm Hg × 3), hyperlipidemia (total cholesterol <200 mg/dl, low-density lipoprotein <120 mg/dl), and impaired fasting glucose or diabetes (fasting glucose <100 mg/dl). A total of 133 eligible Caucasian subjects underwent vascular studies and blood draws for genotyping.

Group 2 was derived from the Morehouse and Emory Team up to Eliminate Health Disparities (META-Health) study, which is a cross-sectional survey of residents in the Atlanta metropolitan area who were invited for assessment of their BP, vascular health, biomarkers, and anthropometric measures, in addition to a series of health questionnaires. These subjects were unselected and recruited by random telephone dialing so as to be representative of the local community. Full details have been published previously.17 History of diabetes, hypertension, cardiovascular disease (coronary artery disease, MI or stroke), and smoking status were defined by participant self report. Subjects of self-reported non-Caucasian race or with documented cardiovascular disease were excluded, allowing inclusion of 270 Caucasian subjects.

Subjects were sampled consecutively and without selection from each study, both of which were approved by the Emory University or Morehouse Institutional Review Boards, and informed consent was obtained from all subjects.

**Central hemodynamic indexes.** All indexes were estimated using noninvasive Sphygmocor technology (Atcor Medical, Sydney, Australia) in a quiet temperature controlled room after an overnight fast. PWA was performed by recording sequential high quality pressure waveforms at the radial artery using a highly sensitive tonometer. The device applies a proprietary transfer function to this peripheral measurement to estimate central (proximal) aortic pressure waveforms. This permits estimation of central (aortic) pressures, pressure augmentation secondary to wave reflections (augmented pressure, AP) and the augmentation index (API/total central pulse pressure, AIx), which is widely considered to be a complex composite measure of wave reflection properties of the arterial tree. Due to its sensitivity to heart rate, a standardized value to 75 bpm (beats per minute) was used for the purposes of this study as advocated by Wilkinson et al.18 Time to reflected wave (TrW) is an indirect measure of stiffness derived from analysis of the pressure wave form and is a measure of the time taken (in ms) for the reflected wave to return from the periphery and merge with the incident wave, where a shorter time indicates faster wave travel, and thus greater arterial stiffness.

Quality control indexes were evaluated at the time of study and non-acceptable readings discarded and repeated. Three readings were taken for each subject and averaged for analysis. Reproducibility studies in our laboratory on nine subjects on consecutive days, by the same operators performing

### Table 1: Characteristics of study populations 1 and 2 (Caucasian subjects)

<table>
<thead>
<tr>
<th>Subject characteristics</th>
<th>PreMed</th>
<th>META-Health</th>
<th>Pooled cohort</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Healthy subjects</td>
<td>Community subjects</td>
<td>n=403</td>
</tr>
<tr>
<td>n</td>
<td>133</td>
<td>270</td>
<td>403</td>
</tr>
<tr>
<td>Age, years</td>
<td>44.8 (14.0)</td>
<td>51.4 (9.1)</td>
<td>49.2 (11.6)</td>
</tr>
<tr>
<td>Male, %</td>
<td>58.9</td>
<td>34.4</td>
<td>44.4</td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>23.1 (2.5)</td>
<td>28.2 (6.6)</td>
<td>26.5 (5.9)</td>
</tr>
<tr>
<td>Brachial systolic BP, mm Hg</td>
<td>116.0 (11.9)</td>
<td>118.5 (15.6)</td>
<td>117.3 (15.1)</td>
</tr>
<tr>
<td>Brachial diastolic BP, mm Hg</td>
<td>68.1 (9.3)</td>
<td>76.1 (10.6)</td>
<td>73.5 (10.8)</td>
</tr>
<tr>
<td>Mean arterial pressure</td>
<td>83.8 (10.4)</td>
<td>91.3 (12.0)</td>
<td>88.2 (11.2)</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>60.5 (9.6)</td>
<td>66.1 (9.7)</td>
<td>59.2 (9.7)</td>
</tr>
<tr>
<td>Total cholesterol, mg/dl</td>
<td>178.9 (26.5)</td>
<td>203.3 (41.9)</td>
<td>194.2 (38.3)</td>
</tr>
<tr>
<td>Glucose, mg/dl</td>
<td>85.9 (8.2)</td>
<td>91.9 (21.4)</td>
<td>90.2 (19.7)</td>
</tr>
<tr>
<td>C-reactive protein, mg/l</td>
<td>1.29 (3.49)</td>
<td>2.6 (3.3)</td>
<td>2.23 (3.6)</td>
</tr>
<tr>
<td>Hypertensive, %</td>
<td>0</td>
<td>31.6</td>
<td>21.1</td>
</tr>
<tr>
<td>Hyperlipidemia, %</td>
<td>0</td>
<td>37.2</td>
<td>24.8</td>
</tr>
<tr>
<td>Diabetic, %</td>
<td>0</td>
<td>5.4</td>
<td>3.7</td>
</tr>
<tr>
<td>Smokers, %</td>
<td>0</td>
<td>13.3</td>
<td>8.9</td>
</tr>
<tr>
<td>Pulse wave analysis indexes</td>
<td>2.9 (5.4)</td>
<td>7.0 (4.6)</td>
<td>5.6 (4.9)</td>
</tr>
<tr>
<td>Augmented pressure, mm Hg</td>
<td>8.5 (14.9)</td>
<td>20.7 (11.3)</td>
<td>16.5 (13.6)</td>
</tr>
<tr>
<td>Time to reflected wave, ms</td>
<td>155.3 (20.5)</td>
<td>144.0 (16.3)</td>
<td>148.6 (18.9)</td>
</tr>
<tr>
<td>Central SBP, mm Hg</td>
<td>102.4 (13.2)</td>
<td>110.9 (15.6)</td>
<td>108.0 (15.5)</td>
</tr>
<tr>
<td>Central DBP, mm Hg</td>
<td>70.0 (9.7)</td>
<td>76.7 (10.8)</td>
<td>74.5 (10.9)</td>
</tr>
</tbody>
</table>

Mean (SD) and percentages are shown. Augmented pressure and index values are heart rate adjusted.

| For between-group comparisons, P < 0.01 for all variables except systolic BP (P = 0.15).
these studies, have demonstrated a coefficient of variation of 20.3 and 2.2% for AIx and TrW respectively. Operators were blinded to genotype results.

**SNP selection and genotyping.** A review of published genome-wide association studies (GWAS) identified variants at 16 loci associating with coronary artery disease and/or MI through June 2009 which had met genome-wide criteria for significance. Of these, three did not replicate in larger follow-up studies (2q36, 15q22, 6q25).19 Another four loci have potential functional relevance to lipid metabolism (1p13–SORT1; 19p13–LDLR; 1p32–PCSK9; 6q26–LpA) and were therefore not included in this study, as we proposed to examine those loci without any obvious mechanism.9,10,20 One single-nucleotide polymorphism (SNP) at 12q24.3 was not included due to its very small effect size in the discovery study.6 Lead SNPs, defined by the original GWAS reports, at the remaining eight loci of interest, which are independent of traditional risk factors, were thus genotyped and are listed in **Supplementary Table S1** online.

Genotyping for both PreMed and META-Health subjects was performed using the SNPstream (Beckman Coulter, Fullerton, CA) 48 plex platform at the Emory University Biomarker Center as part of other candidate gene studies.21,22 The GenomeLab SNPstream Genotyping System Software Suite v2.3 (Beckman Coulter) was used for array imaging and genotype calling. To ensure genotyping accuracy and reproducibility two internal quality control samples were included and each run in triplicate, on each of the 384-well arrays. Genotyping rate for all eight SNPs was >98%.

**Statistical methods.** All continuous variables are described as mean ± s.d., while categorical variables are presented as proportions. The distribution of all variables was tested for normality and none required prior transformation. All SNPs were tested for Hardy–Weinberg equilibrium using PLINK.23 Association tests for quantitative vascular traits were performed using linear regression. SNPs were modeled as independent variables assuming an additive model of inheritance and coded on number of risk alleles as 0, 1 or 2. We tested each SNP for association with the arterial indexes separately in each subject group and then in the total combined group to increase statistical power and reduce type 1 error. Bonferroni adjustment for eight genotypes and five phenotypes \((P \text{ value } \times 13)\) was performed in the pooled analyses, as well as adjustment for age, mean arterial pressure, and height in model 1 which are major determinants of PWA indexes, in addition to gender, glucose, total cholesterol, BMI, and smoking status in model 2.

Two-tailed \(P\) values of <0.05 were considered significant. Bonferroni correction for multiple testing was performed for the eight genotypes and five phenotypes tested \((P \text{ value } \times 13)\). All analyses were performed using PLINK23 and SPSS v17.0 statistical package (SPSS, Chicago, IL).

**Power calculations.** We estimated the effect sizes that would be observed at a fixed power of 80% and an \(\alpha\) of 0.05. Varying the risk allele frequency from the least frequent to most frequent, 0.10–0.45, we estimated that under an additive model, our pooled sample size \((n = 403)\) would allow us to identify effect sizes per risk allele of 2.7–4.4% for AIx; 1.0–1.7 mm Hg for AP, and 3.5–5.5 ms for TrW.

**RESULTS**

Patient characteristics for the Caucasian subjects in PreMed \((n = 133)\) who were healthy and free of all cardiovascular risk factors and for those in META-Health \((n = 270)\) are presented in **Table 1**. Compared to PreMed, subjects in META-Health were older, more likely to be female and almost a third had either hypertension and/or hyperlipidemia, 5% were diabetic, and 13% were current smokers. As expected the central hemodynamic indexes were more impaired for subjects in META-Health compared to those in PreMed (all \(P < 0.01\)) (**Table 1**). Characteristics for the pooled population are also presented in **Table 1**. The distribution of genotypes and allele frequencies in each group for the eight risk variants were in Hardy–Weinberg equilibrium, with allele frequencies similar to published data for Caucasians (**Supplementary Table S1** online).

**Association with vascular traits**

We analyzed the association of these variants with the main quantitative vascular traits, AIx and AP (both standardized to a heart rate of 75 bpm, AIx/AP), TrW, and central SBP and DBP in each study population separately and then combined. The association findings for AIx, AP, and TrW are presented in **Tables 2–4**.

(i) PreMed

The most prominent finding in the PreMed group was the association of the variant at 6p24 with three of the five studied central hemodynamic parameters (**Tables 2–4**). The MI risk allele (C) was associated with worse (higher) AIx \((P < 0.001)\), (higher) AP \((P = 0.001)\), and (lower) TrW \((P = 0.023)\), but not central BP, while the variant at 12q24 showed a borderline trend towards association with AIx \((P = 0.05)\) and AP \((P = 0.09)\) (**Tables 2–4**).

(ii) META-Health

Of the eight SNPs tested in this population, only the 6p24 variant showed significant association with AIx \((P = 0.002)\), AP \((P = 0.024)\), and TrW \((P = 0.037)\) but not central BP, while the variant at 12q24 showed a borderline trend towards association with AIx \((P = 0.05)\) and AP \((P = 0.09)\) (**Tables 2–4**).

(iii) Combined analysis

To minimize a type 1 error we combined the two cohorts for a final analysis. The pooled cohort of 403 subjects showed a significant association between the 6p24 variant and AIx
### Table 2 | Associations between the eight risk variants and augmentation index in the study groups

<table>
<thead>
<tr>
<th>Risk locus</th>
<th>Augmentation index (AIx)</th>
<th>PreMed (n = 133)</th>
<th>META-Health (n = 270)</th>
<th>Pooled (n = 403)</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β (s.e.)</td>
<td>P</td>
<td>β (s.e.)</td>
<td>P</td>
<td>Bonferroni</td>
</tr>
<tr>
<td>1q41</td>
<td>−0.17 (1.8)</td>
<td>0.93</td>
<td>−1.3 (0.9)</td>
<td>0.16</td>
<td>0.50 (0.9)</td>
</tr>
<tr>
<td>2q33</td>
<td>−4.91 (3.4)</td>
<td>0.15</td>
<td>−0.95 (1.5)</td>
<td>0.53</td>
<td>−0.99 (1.6)</td>
</tr>
<tr>
<td>3q22.3</td>
<td>1.09 (2.6)</td>
<td>0.68</td>
<td>0.13 (1.4)</td>
<td>0.93</td>
<td>0.37 (1.4)</td>
</tr>
<tr>
<td>6p24</td>
<td>6.88 (2.0)</td>
<td>&lt;0.001</td>
<td>2.80 (1.0)</td>
<td>0.005</td>
<td>3.50 (1.1)</td>
</tr>
<tr>
<td>9p21</td>
<td>1.40 (1.9)</td>
<td>0.45</td>
<td>−0.52 (1.0)</td>
<td>0.60</td>
<td>0.80 (1.0)</td>
</tr>
<tr>
<td>10q11</td>
<td>1.63 (1.9)</td>
<td>0.40</td>
<td>−0.58 (1.1)</td>
<td>0.59</td>
<td>1.0 (1.0)</td>
</tr>
<tr>
<td>12q24</td>
<td>−3.90 (2.0)</td>
<td>0.05</td>
<td>1.00 (0.9)</td>
<td>0.25</td>
<td>1.41 (1.0)</td>
</tr>
<tr>
<td>21q22</td>
<td>7.30 (2.3)</td>
<td>0.002</td>
<td>1.40 (1.3)</td>
<td>0.31</td>
<td>2.66 (1.3)</td>
</tr>
</tbody>
</table>

The β-coefficients are presented in the direction of the effect of the risk allele (i.e., a positive value indicates a higher level in carriers of the risk allele). Model 1 adjusted for age, MAP, height; Model 2 adjusted for model 1 + gender, cholesterol, glucose, BMI, and smoking.

BMI, body mass index; MAP, mean arterial pressure.

*aBonferroni adjusted for eight variants + five phenotypes.

### Table 3 | Associations between the eight risk variants and augmented pressure in the study groups

<table>
<thead>
<tr>
<th>Risk locus</th>
<th>Augmented pressure (AP)</th>
<th>PreMed (n = 133)</th>
<th>META-Health (n = 270)</th>
<th>Pooled (n = 403)</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β (s.e.)</td>
<td>P</td>
<td>β (s.e.)</td>
<td>P</td>
<td>Bonferroni</td>
</tr>
<tr>
<td>1q41</td>
<td>−0.33 (0.6)</td>
<td>0.61</td>
<td>−0.56 (0.4)</td>
<td>0.12</td>
<td>−0.01 (0.3)</td>
</tr>
<tr>
<td>2q33</td>
<td>−1.41 (1.2)</td>
<td>0.25</td>
<td>−0.51 (0.6)</td>
<td>0.41</td>
<td>−0.43 (0.6)</td>
</tr>
<tr>
<td>3q22.3</td>
<td>0.36 (0.1)</td>
<td>0.71</td>
<td>−0.16 (0.6)</td>
<td>0.78</td>
<td>0.06 (0.5)</td>
</tr>
<tr>
<td>6p24</td>
<td>2.30 (0.7)</td>
<td>0.001</td>
<td>0.80 (0.4)</td>
<td>0.049</td>
<td>1.10 (0.4)</td>
</tr>
<tr>
<td>9p21</td>
<td>0.68 (0.7)</td>
<td>0.30</td>
<td>−0.10 (0.4)</td>
<td>0.83</td>
<td>0.39 (0.4)</td>
</tr>
<tr>
<td>10q11</td>
<td>0.40 (0.7)</td>
<td>0.54</td>
<td>−0.27 (0.4)</td>
<td>0.54</td>
<td>0.22 (0.4)</td>
</tr>
<tr>
<td>12q24</td>
<td>−1.20 (0.7)</td>
<td>0.09</td>
<td>−0.36 (0.4)</td>
<td>0.32</td>
<td>0.39 (0.4)</td>
</tr>
<tr>
<td>21q22</td>
<td>1.95 (0.9)</td>
<td>0.024</td>
<td>0.87 (0.5)</td>
<td>0.10</td>
<td>1.05 (0.5)</td>
</tr>
</tbody>
</table>

The β-coefficients are presented in the direction of the effect of the risk allele (i.e., a positive value indicates a higher level in carriers of the risk allele). Model 1 adjusted for age, MAP, height; Model 2 adjusted for model 1 + gender, cholesterol, glucose, BMI, and smoking.

BMI, body mass index; MAP, mean arterial pressure.

*aBonferroni adjusted for eight variants + five phenotypes.

### Table 4 | Associations between the eight risk variants and time to reflected wave in the study groups

<table>
<thead>
<tr>
<th>Risk locus</th>
<th>Time to reflected wave (TrW)</th>
<th>PreMed (n = 133)</th>
<th>META-Health (n = 270)</th>
<th>Pooled (n = 403)</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β (s.e.)</td>
<td>P</td>
<td>β (s.e.)</td>
<td>P</td>
<td>Bonferroni</td>
</tr>
<tr>
<td>1q41</td>
<td>−3.61 (2.5)</td>
<td>0.14</td>
<td>1.20 (1.3)</td>
<td>0.37</td>
<td>−1.20 (1.2)</td>
</tr>
<tr>
<td>2q33</td>
<td>5.45 (4.7)</td>
<td>0.25</td>
<td>2.40 (2.2)</td>
<td>0.28</td>
<td>2.4 (2.1)</td>
</tr>
<tr>
<td>3q22.3</td>
<td>1.42 (3.5)</td>
<td>0.69</td>
<td>−1.56 (2.1)</td>
<td>0.45</td>
<td>−0.6 (1.9)</td>
</tr>
<tr>
<td>6p24</td>
<td>−6.60 (2.9)</td>
<td>0.023</td>
<td>−3.70 (1.5)</td>
<td>0.013</td>
<td>−4.30 (1.4)</td>
</tr>
<tr>
<td>9p21</td>
<td>−0.60 (2.6)</td>
<td>0.82</td>
<td>−0.23 (1.4)</td>
<td>0.87</td>
<td>−0.89 (1.3)</td>
</tr>
<tr>
<td>10q11</td>
<td>−3.90 (2.6)</td>
<td>0.15</td>
<td>0.95 (1.6)</td>
<td>0.55</td>
<td>−1.17 (1.4)</td>
</tr>
<tr>
<td>12q24</td>
<td>2.20 (2.8)</td>
<td>0.43</td>
<td>−1.30 (1.3)</td>
<td>0.32</td>
<td>0.52 (1.3)</td>
</tr>
<tr>
<td>21q22</td>
<td>−7.10 (3.3)</td>
<td>0.037</td>
<td>−3.30 (1.9)</td>
<td>0.10</td>
<td>−4.11 (1.7)</td>
</tr>
</tbody>
</table>

The β-coefficients are presented in the direction of the effect of the risk allele (i.e., a positive value indicates a higher level in carriers of the risk allele). Model 1 adjusted for age, MAP, height; Model 2 adjusted for model 1 + gender, cholesterol, glucose, BMI, and smoking.

BMI, body mass index; MAP, mean arterial pressure.

*aBonferroni adjusted for eight variants + five phenotypes.
homozygotes having an average value of 18.4% compared to the sample. The association with AIx appeared most robust, with replicated in a larger community population a healthy cohort free from confounding risk factors or medi-
cations, and (rs12526453) was associated with greater AIx, AP, and TrW in the arterial tree in two independent cohorts.

gene, identified through GWAS of early onset MI, is significant-
that the MI risk locus at chromosome 6p24 in the PHACTR1
ies, all of which could be considered as representing small ves-
el or systemic arterial stiffness. Furthermore, the TrW is also considered a surrogate of large artery stiffness. Importantly
ormalities in central hemodynamic indexes including AIx are powerful predictors of cardiovascular morbidity and mortality. A recent meta-analysis of population cohorts confirms that a 10% rise in AIx confers ~30% increase in risk for all cause mortality. Thus, the independent association of this 6p24 variant with several related central hemodynamic indexes suggests that it may mediate risk of MI by modulating arterial elastic properties and pressure wave reflections.

Of the remaining variants tested for association in the larger pooled cohort (n = 403) no further significant associations were identified with any phenotype (Tables 2–4).

**DISCUSSION**

In order to explore potential mechanistic pathways of newly identified genetic variants associating with MI, whose function is currently unknown, we investigated their relationship with the intermediate phenotype of PWA indexes. We demonstrate that the MI risk locus at chromosome 6p24 in the PHACTR1 gene, identified through GWAS of early onset MI, is significantly associated with abnormal wave reflection properties of the arterial tree in two independent cohorts.

In this study, the risk allele (C) of the 6p24 variant (rs12526453) was associated with greater AIx, AP, and TrW in a healthy cohort free from confounding risk factors or medications, and replicated in a larger community population sample. The association with AIx appeared most robust, with homozygotes having an average value of 18.6% compared to the referent group mean of 12.7%. AIx is considered a complex measure of wave reflections and elasticity of the muscular arteries, all of which could be considered as representing small vessel or systemic arterial stiffness. Furthermore, the TrW is also considered a surrogate of large artery stiffness. Importantly abnormalities in central hemodynamic indexes including AIx are powerful predictors of cardiovascular morbidity and mortality. A recent meta-analysis of population cohorts confirms that a 10% rise in AIx confers ~30% increase in risk for all cause mortality. Thus, the independent association of this 6p24 variant with several related central hemodynamic indexes suggests that it may mediate risk of MI by modulating arterial elastic properties and pressure wave reflections.

Interestingly, we did not observe any association with central SBP or DBP in either study cohort. This is perhaps not surprising, given that AIx has been shown to be considerably more heritable than BP. For example in a study of twins, Snieder et al. demonstrated a heritability of 37% for AIx, but only 13–25% for BP traits, while others have found estimates as high as 62% for AIx. This is likely due to the fact that arterial elastic properties are just one determinant of BP and genetic variants are more likely to explain greater variance in these upstream components than in the final composite phenotype.

Although there were promising initial associations between at least two other SNPs at 12q24 and 21q22 with these parameters, none were successfully replicated in the second cohort with risk factors. Other variants did not associate significantly with any phenotype although this could be explained by lack of statistical power to detect much smaller effect sizes.

The rs12526453 SNP resides on the short arm of chromo-
some 6, in the untranslated portion of the PHACTR1 gene but is in linkage disequilibrium with several neighboring SNPs within this gene (Supplementary Figure S1 online). PHACTR1 codes for phosphatase and actin regulator 1, which is one of many regulating proteins for the protein phosphatase 1 (PP1) enzyme. The latter is a ubiquitously expressed enzyme involved in a wide array of physiological processes, including gene expression, muscle contraction, and glycogen metabolism as well as being a critical negative regulator of Ca2+ cycling and contractility in smooth muscle cells and cardiomyocytes, which may be pertinent in the context of vaso-

**Figure 1** | Mean augmentation index (HR 75 standardized) per risk allele at 6p24, represented by SNP rs12526453 (risk allele C). Values are adjusted for mean arterial blood pressure and age. Standard error bars shown. HR, heart rate, SNP, single-nucleotide polymorphism.
In conclusion, we have shown that the 6p24 marker, rs12526453, which was recently identified as conferring risk of MI through genome-wide association, also associates with impaired central hemodynamic indexes in two distinct populations independent of age and BP. This finding could have implications for understanding the mechanism of risk for this locus as well as opening up possible therapeutic interventions in those with the risk allele.

**Limitations**

Limitations of our study include its cross-sectional design which restricts the conclusions we can draw regarding causality, although refining the phenotype in this manner provides some insight into potential risk pathways. Second, our restricted sample size meant that we may have missed SNP effects of a smaller magnitude than our power calculations suggested or those that have interactions with environmental factors. While we combined cohorts to maximize power in the pooled analysis, we accept that the significant baseline differences in the characteristics of the cohorts may have introduced some heterogeneity and bias. We were also unable to adjust for other possible confounders which could influence the studied outcomes such as menopause status of female participants or specific medication effects for those in the META-Health group with risk factors. On the other hand the nature of our study necessitates multiple testing, which could lead to false-positive findings. However, some degree of confidence in our findings is provided by their persistence despite conservative Bonferroni adjustment, as well as the fact that the association with 6p24 was with several linked parameters in two distinct populations which was apparent even with relatively small sample sizes. Finally, we did not have sufficient data to examine association with pulse wave velocity, an independent and gold standard measure of large artery stiffness, to differentiate if the observed effects were due primarily to enhanced stiffness.30

**Implications**

GWAS have identified multiple variants associating with MI, but mechanisms of risk remain poorly understood. For 6p24, if MI risk is confirmed to be mediated via impaired arterial elastic properties then this could have significant treatment implications. First, existing therapies known to modulate wave reflections could be used to try and ameliorate risk in those with the 6p24 variant in a personalized genotype-targeted manner. Second, new treatments targeting the PHACTR1 gene, modulating calcium regulation may offer a new approach to reducing wave reflections and reducing risk associated with pressure augmentation in the broader population.

Supplementary material is linked to the online version of the paper at http://www.nature.com/ajh

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