ORIGINAL ARTICLE

Genetic variation in the alpha1B-adrenergic receptor and vascular response

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INTRODUCTION

Alpha1-adrenergic receptors (α1-ARs) are the prime mediators of vasoconstriction induced by activation of the sympathetic nervous system and thus have an important role in blood pressure regulation. Among the three α1-AR subtypes (α1A-, α1B- and α1D- AR), the α1A-AR appears to be the principal mediator of physiological vasoconstriction,1,2 while the α1B- and α1D-AR also contribute, as demonstrated in various human and animal models.3,4

The association between genetic variants in the α1A-AR gene (ADRA1A) with outcomes such as vasoconstriction and blood pressure has been studied in a number of experimental models. In a human hand vein model, variation in ADRA1A did not explain the large interindividual variability or the ethnic differences in vasoconstrictor responses to the infusion of the selective α1-AR agonist, phenylephrine.5-7 Also, ADRA1A variation was not associated with the rise in blood pressure after sympathetic stimulation in the setting of experimental stress.8-10

However, a recent study in 1614 Nigerians examining common variants in candidate genes of 28 different pathways implicated in the alpha1-adrenergic pathway.11 Among the α1-AR-pathway genes, variants in the α1B-AR were most significantly associated with blood pressure and hypertension, while variants in the α1A-AR were not, suggesting that genetic variants in the α1B-AR, rather than in the α1A-AR subtype, may contribute to interindividual variability in the regulation of vascular tone and blood pressure control.

α1B-ARs contribute to vasoconstriction in animal studies, possibly by mediating smooth muscle contraction directly and also by regulating the expression and function of other adrenergic receptors.12-14 Little is known about the effects of ADRA1B variation on vascular responses in humans. An early study found no association between phenylephrine-mediated vasoconstriction and four infrequent ADRA1B coding variants in 45 subjects with and without hypertension.15 Subsequently, the genetic architecture of ADRA1B was explored systematically in various ethnic groups, showing great interindividual and interethnic variability.16

We therefore set out to define the association between ADRA1B genotypes and vascular sensitivity to vasoconstriction induced by an α1-agonist using two experimental models: local venous stimulation in the dorsal hand vein, and the increase in blood pressure during the cold pressor test (CPT), reflecting the systemic cardiovascular response to acute sympathetic activation. Previous studies showed ethnic differences in these responses, with African Americans having a greater blood pressure increase after cold pressor stress and greater sensitivity to phenylephrine-induced vasoconstriction compared with Caucasians, suggesting that genetic factors may contribute to these responses.5,17-18 Thus, we tested the hypothesis that variation in ADRA1B contributes to interindividual and ethnic differences in agonist-mediated vasoconstriction and in the stress-induced increase in blood pressure following a CPT.

MATERIALS AND METHODS

Subjects

We studied dorsal hand vein responses in 105 healthy normotensive Caucasians and African Americans aged 18–45 years. Details of the study procedures and analyses of other genes were published previously.5,7

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The alpha1B (α1B)-adrenergic receptors contribute to vasoconstriction in humans. We tested the hypothesis that variation in the ADRA1B gene contributes to interindividual variability and ethnic differences in adrenergic vasoconstriction. We measured dorsal hand vein responses to increasing doses of phenylephrine in 64 Caucasians and 41 African Americans and genotyped 34 ADRA1B variants. We validated findings in another model of catecholamine-induced vasoconstriction, the increase in mean arterial pressure (ΔMAP) during a cold pressor test (CPT). One ADRA1B variant, rs10070745, present in 14 African-American heterozygotes but not in Caucasians, was associated with a lower phenylephrine ED50 (geometric mean (95% confidence interval), 144 (69–299) ng ml−1) compared with 27 African-American non-carriers (208 (130–334) ng ml−1; P = 0.015) and contributed to the ethnic differences in ED50. The same variant was also associated with a greater ΔMAP during CPT (P = 0.008). In conclusion, ADRA1B rs10070745 was significantly associated with vasoconstrictor responses after adrenergic stimulation and contributed to the ethnic difference in phenylephrine sensitivity.
Fifty-seven of the 105 subjects also participated in a second study that included a CPT. Pregnant females were excluded, and subjects took no medications for at least 2 weeks, abstained from alcohol and caffeine for at least 5 days, and received a diet containing 150 mmol per day of sodium, 70 mmol per day of potassium, and 600 mmol per day of calcium for at least 4 days before each study day. The Institutional Review Board of Vanderbilt University Medical Center approved the study protocols, and all the subjects provided written informed consent.

Venous response to phenylephrine
Venous responses were measured in a dorsal hand vein with a linear variable differential transformer, as previously described. In summary, a 24-gauge intravenous cannula was inserted into a suitable right dorsal hand vein and kept patent with saline solution infused at a flow rate of 0.4 ml min$^{-1}$. A linear variable differential transformer (MHR 100; Shaevitz Engineering, Pennsaken, NJ, USA) was mounted on the dorsum of the subject's hand. A second intravenous cannula was inserted for blood sampling into the antecubital vein of the contralateral arm. After 30 min of saline infusion, a blood sample was taken for the determination of baseline plasma catecholamines and for genotyping. We determined the baseline vein diameter while a sphygmomanometer cuff around the upper arm was inflated to 50 mm Hg to induce venous filling. After three stable baseline measurements, we assessed vein constriction in response to increasing doses of phenylephrine, an $\alpha_1$-AR agonist. Phenylephrine (Eliksin-Sinn, Cherry Hill, NJ, USA) was infused through the cannula with a syringe infusion pump (Harvard Apparatus, Holliston, MA, USA) at increasing dose rates (range, 12–12 000 ng min$^{-1}$). The infusion at each dose rate lasted 7 min, and the vein diameter was measured during the last 2 min of the infusion. The total flow rate through the vein was kept constant at 0.4 ml min$^{-1}$ throughout the various phenylephrine dilutions. The heart rate and blood pressure were continuously monitored with a bedside cardiac monitor (Dinamap MPS; Johnson and Johnson Medical, Tampa, FL, USA).

Analysis of hand vein response to phenylephrine
Venous constriction was expressed as the percentage reduction in vein diameter from average baseline measurements, plotted against increasing doses of phenylephrine in individual semi-logarithm dose–response graphs and analyzed using a sigmoid dose–response model with variable slope (GraphPad Prism 4.03, GraphPad, La Jolla, CA, USA). We determined the phenylephrine dose that produced 50% of maximal constriction (ED$_{50}$, representing sensitivity to the drug) and also calculated the maximal venoconstriction ($E_{\text{max}}$, representing maximum response) for each subject. The analyses were performed by a single investigator unaware of the subject's genotype.

Cold pressor test
The CPTs were performed as previously described. All the preparations for the CPT were performed only after the resting measurements had been obtained and after 30 min supine rest to minimize confounding through anticipation. With the subject in a supine position, the left foot was fully immersed up to the ankle for 2 min in a tub filled with a slurry of ice and water (4 °C). Two readings of the blood pressure and heart rate were taken with the semi-automated device (Dinamap MPS; Johnson and Johnson Medical, Tampa, FL, USA), starting at approximately 15 and 45 s after foot immersion. At 1 min, a blood sample (10 ml) was taken for the determination of plasma catecholamine concentrations.

Determination of plasma catecholamine concentrations
The blood was collected into cooled heparinized tubes that were placed on ice until centrifuged at 4 °C for 10 min at 3000 r.p.m. The plasma was separated and stored at −20 °C in previously cooled tubes containing 40 μl of reduced glutathione (6%) until assayed. Noradrenaline and epinephrine concentrations were measured by high-performance liquid chromatography using electrochemical detection with dihydroxybenzylamine as internal standard.

Genotyping
We genotyped 34 ADRB1 SNPs listed in Supplementary Table S1. We selected 24 tagSNPs for ADRB1 (chromosome 5, position 159269–159343 kb) from the Hapmap project using Haploview 4.2 software.

Using data based on Utah residents with Northern and Western ancestry (CEU) and on subjects of African ancestry in southwest USA (ASW). We excluded individuals from the Hapmap project with more than 5% missing genotypes and SNPs with minor allele frequencies less than 5%. Two-marker haplotypes were used to tag SNPs in strong linkage disequilibrium, defined as $r^2 > 0.8$. Furthermore, we included five previously published tag SNPs that were not captured in the Haploview tagging, and five additional SNPs previously associated with clinical outcomes (Supplementary Table S1). Genotyping was performed using the Sequenom platform (MassArray, San Diego, CA, USA). For quality control, we included negative and positive controls with each genotyping run. Quality-control procedures included examination of marker and sample genotyping efficiency, allele-frequency calculations, and tests of Hardy–Weinberg equilibrium.

Statistical analysis
The cohort was a convenience sample consisting of participants in previous studies, and the sample size was not based on a priori calculation. The primary outcome for the hand vein study was drug sensitivity (expressed as ED$_{50}$), and drug efficacy ($E_{\text{max}}$) was the secondary outcome. ED$_{50}$ values were not normally distributed and were therefore log transformed and expressed as geometric means with 95% confidence intervals (CIs). The primary outcome for the CPT was the change in mean arterial blood pressure (DMax). We compared the outcomes among genotypes in a single marker analysis first in each ethnic group separately and then in the combined cohort. We then adjusted for potential confounders that were associated with the outcomes in our previous studies using the same endpoints. For the hand vein study, we adjusted for sex, BMI, resting norepinephrine concentrations and, for analyses of the combined cohort, ethnicity, using a multiple linear regression model. The secondary outcome, phenylephrine efficacy ($E_{\text{max}}$), was compared among genotypes using the non-parametric Kruskal–Wallis test. SNPs that were nominally significant in the hand vein study for either ED$_{50}$ or $E_{\text{max}}$ were then tested for association with blood pressure response during the CPT for validation. For the CPT, we adjusted for age, sex, BMI and baseline mean arterial blood pressure, and additionally for ethnicity in analyses of the combined cohort.

For all the genetic analyses, we assumed an additive genetic model, coding the genotypes according to the number of variant alleles (0–2). We used PLINK software (v. 1.07) to assess overall differences in the outcomes among the genotypes. Other statistical analyses were performed using SPSS software (v. 21, IBM SPSS, Chicago, IL, USA). All the analyses were two-tailed, and a $P$-value < 0.05 was considered significant; permutation tests were performed to ensure that the empirical $P$-value of all SNPs was also significant at the 0.05 threshold.

RESULTS
Genotyping
Minor allele frequencies for 34 ADRB1 SNPs in 105 subjects were in the expected range, and all genotypes conformed to Hardy–Weinberg equilibrium (Supplementary Table S1). We did not identify any carriers of the rs10070745, rs7736470 and rs876529 variant in Caucasians.

Hand vein study

Subjects and outcomes. African Americans (n = 41) had a higher BMI (P = 0.044), diastolic blood pressure (P = 0.034) and a higher resting heart rate (P = 0.004) than Caucasians (n = 64). There was wide interindividual variability in response to phenylephrine, with the range of ED$_{50}$ spanning three log units (11 to 5442 ng min$^{-1}$), geometric mean, 260 ng min$^{-1}$; 95% CI: 202 to 335 ng min$^{-1}$; Table 1), and the $E_{\text{max}}$, ranging from 13.7% to 100% (median, 87%; interquartile range, 76% to 97%; Table 1). As previously reported in this cohort, African Americans had a lower ED$_{50}$ (that is, greater sensitivity; adjusted $P = 0.006$) and a trend to a higher $E_{\text{max}}$ compared with Caucasians ($P = 0.079$).

ADRB1 variants and phenylephrine response. Among the 34 ADRB1 variants, one variant (rs10070745) was associated with the primary outcome, phenylephrine sensitivity (Table 2). The
Table 1. Demographic and cardiovascular measurements in subjects who performed the hand vein study \((n = 105)\) and the subgroup that also participated in the cold pressor test \((n = 57)\)

| Characteristics                  | Hand vein study, \(n = 105\) | Cold pressor test, \(n = 57\) |
|----------------------------------|-------------------------------|-------------------------------|
| Age, years                       | 27.3 ± 7.2                    | 26.4 ± 5.8                    |
| Female sex, \(n\) (\%)           | 47 (44.8)                     | 23 (40)                       |
| Caucasians, \(n\) (%)            | 64 (61)                       | 36 (63)                       |
| BMI, kg m\(^{-2}\)               | 25.3 ± 4.5                    | 24.8 ± 4.1                    |
| Resting SBP, mm Hg               | 111.6 ± 11.5                  | 115.5 ± 10.6                  |
| Resting DBP, mm Hg               | 62.4 ± 8.3                    | 64.5 ± 6.3                    |
| Resting MAP, mm Hg               | 72.1 ± 6.6                    | 81.5 ± 6.8                    |
| Resting HR, b.p.m.               | 59.9 ± 8.3                    | 64.0 ± 7.1                    |
| Baseline plasma norepinephrine, pg ml\(^{-1}\) | 8.3 ± 6.6                     | 9.1 ± 9.1                     |
| Phenylephrine ED\(_{50}\) (ng min\(^{-1}\)), geometric mean (95% CI) | 167.0 (76–97) | 193.6 (72–95) |
| Phenylephrine \(E_{\text{max}}\) %, median (IQR) | 87 (76–97) | 82 (72–95) |
| \(\Delta\text{MAP}\) after CPT, mm Hg | 13.3 (10–16) | 14.9 ± 13.4 |
| \(\Delta\text{HR}\) after CPT, b.p.m. | 14.5 (12–19) | 14.9 ± 13.4 |

Abbreviations: BMI, body mass index; b.p.m., beats per minute; CPT, cold pressor test; DBP, diastolic blood pressure; HR, heart rate; IQR, interquartile range; MAP, mean arterial pressure; SBP, systolic blood pressure.

Table 2. Genetic variants in ADRA1B associated with sensitivity to phenylephrine (ED\(_{50}\))

| SNP          | Caucasians | African Americans | P-value* |
|--------------|------------|-------------------|----------|
|              | \(ED_{50}\) (ng min\(^{-1}\)), geometric mean (95% CI) | \(ED_{50}\) (ng min\(^{-1}\)), geometric mean (95% CI) | |
| Number of variant alleles | 0 | 1 | 2 | 0 | 1 | 2 |
| rs10070745   | 325 (234–452) | 208 (130–334) | 0.015 |
| \(n = 64\)   | \(n = 0\) | \(n = 0\) | |
| rs7737796    | 195 ± 13.8 | 19.5 ± 13.8 | 0.89    |
| \(n = 64\)   | \(n = 0\) | \(n = 0\) | |

Abbreviations: CI, confidence interval; SNP, single-nucleotide polymorphism. *P-value adjusted for sex, body mass index and baseline norepinephrine.

rs10070745 variant, present in 14 African-American heterozygotes but not in Caucasians, was associated with lower \(ED_{50}\) (β-coefficient = −0.47; 95% CI: −0.84 to −0.10; adjusted \(P = 0.015\), Figure 1). This variant also contributed to the ethnic differences in \(ED_{50}\) the effect of ethnicity on \(ED_{50}\) (β = −0.29; 95% CI: −0.59 to −0.10, \(P = 0.006\)) was weakened and no longer statistically significant after adding the rs10070745 genotype to the adjusted model in the combined cohort (β = −0.20; 95% CI: −0.54 to 0.05, \(P = 0.11\)).

The second outcome, phenylephrine \(E_{\text{max}}\) was marginally associated with three variants (Table 3). The rs952037 variant was associated with higher \(E_{\text{max}}\) (higher efficacy) in the combined group (\(P = 0.041\)). The rs7737796 variant showed a nonsignificant trend to decreased \(E_{\text{max}}\) in both ethnic groups, which was statistically significant in the combined group (\(P = 0.018\), Table 3). The rs17057303 showed a borderline association with lower \(E_{\text{max}}\) in African Americans (\(P = 0.044\), Table 3).

Cold pressor test

Subjects and outcomes. Of the 105 subjects that completed the hand vein study, 57 also participated in the CPT study. The baseline demographics of this subgroup were similar to those of the whole group (Table 1). Following CPT, there was a significant increase in systolic blood pressure (ΔSBP), diastolic blood pressure (ΔDBP) and mean arterial pressure (ΔMAP; all \(P < 0.001\)), without ethnic differences in these responses (all \(P > 0.22\)).

ADRA1B variants and blood pressure response to CPT. Of the four SNPs that were significantly associated with outcomes in the hand vein test, two SNPs (rs10070745 and rs17057303) were also associated with ΔMAP during the CPT (Table 4). The eight African-American subjects carrying the rs10070745 minor allele, the only variant associated with the primary outcome in the hand vein model (greater sensitivity to phenylephrine), had a 29% greater mean ΔMAP (22 mm Hg; 95% CI: 14–30 mm Hg) compared with the 13 African-American non-carriers (mean ΔMAP, 17 mm Hg; 95% CI: 11–23 mm Hg; \(P = 0.008\), Table 4, Figure 2). Furthermore, rs17057303, which was associated with a lower \(E_{\text{max}}\) in the hand vein study among African Americans, also showed a trend to a lower ΔMAP following the CPT among African Americans and in the combined cohort (Table 4). In the combined cohort, the four heterozygotes had a 44% lower mean ΔMAP (10 mm Hg; 95% CI: 2–19 mm Hg) compared with the 53 non-carriers (18 mm Hg; 95% CI: 16–21 mm Hg; \(P = 0.050\)).

DISCUSSION

The major new finding of the study is that genetic variation in ADRA1B affects phenylephrine-mediated venoconstriction and...
blood pressure changes following CPT. One variant, rs10070745, present only in African Americans, was associated with both phenylephrine-mediated venoconstriction and blood pressure increase during the CPT.

Vascular α1B-ARs are expressed in arterial and venous beds.1,4,25 The α1B-AR appears to mediate vasoconstriction by directly activating the Gq signaling pathway, leading to increased intracellular calcium through generation of the second messenger inositol (1,4,5)-triphosphate and diacylglycerol, and by regulating the expression and function of other adrenergic receptors, especially α1D-ARs.3,26,27 Animal studies using α1B-AR knockout mice revealed that the blood pressure response to phenylephrine was decreased by 45% compared with the wild type, suggesting that α1B-ARs have an important role in vascular smooth muscle contraction and blood pressure changes in response to an α1-AR agonist.12

ADRA1B consists of two exons separated by a 20-kb intron and is located in a locus containing important candidate genes for blood pressure regulation.28 Genome-wide analyses have generally not found an association between ADRA1B and hypertension or resting blood pressure measurements.11,29 On the other hand, a recent candidate gene study in a Nigerian population found the strongest association between both blood pressure and hypertension with variants in ADRA1B.11 However, resting blood pressure is a phenotype affected by many factors. Therefore, to more specifically address the functional effects of ADRA1B variants, we used two models of adrenergically mediated vasoconstriction, the dorsal hand vein response to phenylephrine and the increase in blood pressure in response to sympathetic activation induced by a cold stimulus.

Vascular studies performed in the human dorsal hand vein provide several advantages. Most important, low doses of agonist that have minimal systemic effects are infused, and thus measures of vascular sensitivity can be obtained in vivo without the reflex responses that accompany systemic infusions of vasoactive drugs. Many investigators reported great interindividual variation in phenylephrine-mediated venoconstriction; much of this variability is thought to be genetic.30,31 However, the genetic determinants of variability in α1-AR-mediated vascular responses have not been elucidated.

We previously reported increased sensitivity (lower ED50) for venous and arterial α1-AR-mediated vasoconstriction in African Americans compared with Caucasians.5,7 However, α1-AR-mediated vasoconstriction was similar in the two groups, suggesting that ethnic differences in α1-ARs and their proximal signal transduction pathway (for example, coupling, phospholipase C activation, calcium release from sarcoplasmatic reticulum) could explain the ethnic differences in α1-AR-mediated vasoconstriction.32 On the basis of this premise, we previously studied ADRA1A variants and found that they did not explain the ethnic differences in phenylephrine sensitivity, although two ADRA1A variants explained some of the interindividual variation.7 In the present study, the ADRA1B variant, rs10070745, was

![Figure 1. ADRA1B rs10070745 association with sensitivity to phenylephrine (ED50) in African Americans. The columns show geometric means, the error bars the 95% confidence intervals. Carriers of the variant allele had a significantly lower geometric mean ED50 (P = 0.015).](image)

Table 3. Genetic variants of ADRA1B associated with maximum response to phenylephrine (E_{max})

| SNP          | Caucasians | African Americans | All     |
|--------------|------------|-------------------|---------|
|              | E_{max} (%), median (IQR) | P-value | E_{max} (%), median (IQR) | P-value | P-value |
|              | Number of variant alleles |         | Number of variant alleles |         |         |
| rs952037     | 82 (66–93) | 85 (75–97) | 92 (72–100) | 0.39 | 87 (73–96) | 90 (82–98) | 94 (87–100) | 0.24 | 0.041 |
|              | n = 29 | n = 31 | n = 4 | | n = 11 | n = 19 | n = 11 | | |
| rs7737796    | 92 (82–98) | 79 (64–88) | 82 (66–99) | 0.053 | 95 (86–98) | 86 (76–94) | 98 (87–100) | 0.10 | 0.018 |
|              | n = 24 | n = 32 | n = 8 | | n = 10 | n = 24 | n = 7 | | |
| rs17057303   | 85 (75–96) | 61 | | 0.20 | 92 (82–99) | 85 (64–89) | | 0.044 | 0.11 |
|              | n = 63 | n = 1 | n = 0 | | n = 34 | n = 7 | n = 0 | | |

Abbreviation: IQR, interquartile range; SNP, single-nucleotide polymorphism.
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Dorsal hand vein responses represent a challenging phenotype that makes replication in a second cohort unfeasible. Thus, we examined associations of candidate variants identified in the hand vein model in a second vascular phenotype in which α1-AR-mediated vasoconstriction has a role. The CPT causes substantial sympathetic activation and vasoconstriction leading to an increase in blood pressure. Concordant with increased sensitivity to phenylephrine in the dorsal hand vein, subjects with rs10070745 had a greater rise in mean arterial pressure following the CPT, suggesting greater vasoconstriction. The finding that the same variant is associated with two related vasoconstrictor phenotypes supports the validity of the findings.

In keeping with published data, the intronic rs10070745 variant was present in 17% of African Americans but not in Caucasians. We did not find a report of an association of this variant with any biological function. However, in the candidate gene study in a large Nigerian population that implicated ADRA1B variants on arterial vasoconstriction. However, there is no selective α1B-AR agonist available for use in humans, and phenylephrine is not known to have affinity for other adrenergic receptors in the human vascular beds. Our findings were derived using the dorsal hand vein model and may therefore not automatically be extrapolated to other venous or arterial vascular beds. However, we previously found similar ethnic differences in α1-AR-mediated vasoconstriction in both venous and arterial vascular beds, suggesting that responsiveness in the dorsal hand vein model may also reflect that in arterial vascular beds. It will be interesting to study the effects of the ADRA1B variants on arterial vasoconstriction. However, these studies are invasive and therefore difficult to conduct. Furthermore, although our sample size for the dorsal hand vein model study was fairly large for a translational study, some genotype groups were small, and we did not account for the multiple comparisons required by the large number of ADRA1B alleles.
variants in the first study. Finally, our sample was not large enough to include homozygotes for the rs10070745 variant. Thus, our findings regarding the association of the rs10070745 variant with responsiveness to phenylephrine and CPT are hypothesis-generating and need to be validated in other populations.

In conclusion, we found that the ADRA1B rs10070745 variant, present in African Americans only, was associated with increased sensitivity to phenylephrine-mediated venoconstriction, contributed to the ethnic differences in hand vein response, and was associated with higher blood pressure responses to a CPT. Further studies exploring the association of this variant with blood pressure phenotypes and hemodynamic responsiveness in black populations, and particularly in subjects homozygous for the variant, will be of interest.

CONFLICT OF INTEREST
The authors declare no conflict of interest.

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REFERENCES
1 Rudner XL, Berkowitz DE, Booth JV, Funk BL, Cozart KL, D’Arminco EB et al. Subtype specific regulation of human vascular alpha(1)-adrenergic receptors by vessel bed and age. Circulation 1999; 100: 2336–2343.
2 Leech CJ, Faber JE. Different alpha-adrenoceptor subtypes mediate constriction of arterioles and venules. Am J Physiol 1996; 270: H710–H722.
3 Docherty JR. Subtypes of functional alpha1-adrenoceptor. Cell Mol Life Sci 2010; 67: 405–417.
4 Yan M, Sun J, Bird PI, Liu DL, Grigg M, Lim YL. Alpha1A- and alpha1B-adrenoceptors are the major subtypes in human saphenous vein. Life Sci 2001; 68: 1191–1198.
5 Adeefurin A, Ghimire LV, Kohli U, Muszkat M, Sofowora GG, Paranjape SY et al. Blacks have a greater sensitivity to alpha1-adrenoceptor-mediated venoconstriction compared with Whites. Hypertension 2013; 61: 915–920.
6 Sofowora GG, Dishy V, Landau R, Xie HG, Prasad HC, Byrne DW et al. Alpha 1A-adrenergic receptor polymorphism and vascular response. Clin Pharmacol Ther 2004; 75: 539–545.
7 Adeefurin A, Ghimire LV, Kohli U, Muszkat M, Sofowora GG, Li C et al. Genetic variation in the alpha-adrenergic receptor and phenylephrine-mediated venoconstriction. Pharmacogenomics J 2014; 15: 310–315.
8 Kelsey RM, Alpert BS, Dahmer MK, Krushkal J, Quasney MW. Alpha-adrenergic receptor gene polymorphisms and cardiovascular reactivity to stress in Black adolescents and young adults. Psychophysiology 2012; 49: 401–412.
9 Yang X, Gu D, He J, Hixon JE, Rao DC, Lu F et al. Genome-wide linkage and regional association study of blood pressure response to the cold pressor test in Han Chinese: the genetic epidemiology network of salt sensitivity study. Circ Cardiovasc Genet 2014; 7: 521–528.
10 He J, Kelly TN, Zhao Q, Li H, Huang J, Wang L et al. Genome-wide association study identifies 8 novel loci associated with blood pressure responses to interventions in Han Chinese. Circ Cardiovasc Genet 2013; 6: 598–607.
11 Reder NP, Tayo BO, Salako B, Ogunniyi A, Adeyemo A, Rotimi C et al. Adrenergic alpha-1 pathway is associated with hypertension among Nigerians in a pathway-focused analysis. PLoS One 2012; 7: e37145.