

Variability in β -adrenergic receptor response in the vasculature: Role of receptor polymorphism

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β -adrenergic receptors (β ARs) are important in determining vascular tone. Stimulation of β_2 ARs in the vasculature produces vasodilatation, which acts to counteract the vasoconstriction produced by endogenous sympathetic agonists. Impaired vasodilatation occurs in human hypertension and in Blacks. It is now clear that receptor polymorphism may contribute to some of the variability seen in vascular response. β_2 AR haplotypes determine the desensitization response to chronic agonist exposure in vivo and result in altered vasodilatory responses. (*J Allergy Clin Immunol* 2002;110:S318-21.)

Key words: β -adrenergic receptors, vascular response, pharmacogenetics, hypertension, ethnicity

In addition to determining the response to both physiologic and pharmacologic stimuli in the pulmonary system, β -adrenergic receptors (β ARs) are critical to normal homeostasis in the cardiovascular system. Stimulation of β_2 ARs in the vasculature produces vasodilatation, whereas stimulation of β_1 ARs on the heart has positive chronotropic effects. There is considerable variability in the response to endogenous and exogenous agonists, both between healthy persons of different ethnic groups and between healthy persons and persons with diseases such as hypertension.

β AR-MEDIATED VASODILATATION IN HYPERTENSION

The study of β AR response in the vasculature is difficult because of the complicating effects of cardiovascular reflexes, which are stimulated by changes in vascular tone produced by the administration of vasoactive agents. For that reason vascular responses are best studied after direct infusion of agonists into the vascular bed of interest at doses insufficient to produce systemic effects. By using the technique of infusion of isoproterenol directly into the brachial artery of persons with hypertension, we and others have studied vascular response in the human forearm.¹ By using this technique it was clear (Fig 1) that the increase in forearm blood flow was significantly lower in hypertensive than normotensive persons, imply-

Abbreviations used

ANOVA: Analysis of variance

β AR: β -adrenergic receptor

ing that in hypertensive persons, the ability to vasodilate in response to β_2 -adrenergic stimulation is impaired.¹ This impairment was not explained by altered basal or stimulated sympathetic activity.¹

β AR-MEDIATED VASODILATATION IN DIFFERENT ETHNIC GROUPS

There are marked racial differences in the frequency and severity of hypertension, with blacks being at particular risk for hypertension. Even when matched with other ethnic groups for the severity of hypertension, blacks appear to have increased rates of morbidity associated with hypertension. The increase in blood pressure is associated with increased peripheral vascular resistance, which in turn reflects both impaired vasodilatation and increased vasoconstriction. By using the technique mentioned previously, direct intrabrachial artery infusion of isoproterenol and measurement of the increase in forearm blood flow, we demonstrated a marked impairment of β_2 AR-mediated vasodilatation in blacks.² The forearm blood flow in whites increased almost 10-fold compared with a less than 4-fold increase in blacks (Fig 2). The impaired vasodilatation in blacks is accompanied by increased α -adrenergically mediated vasoconstriction,³ and the combination of these vascular changes results in additive effects in response to endogenously released catecholamines that act as mixed β - and α -agonists (Fig 3).

ROLE OF β AR POLYMORPHISMS IN HYPERTENSION

The recognition that polymorphisms of the β_2 AR occur frequently led to the obvious question of whether such polymorphisms are associated with hypertension. In an Afro-Caribbean population, the Gly16 polymorphism was said to occur more frequently in hypertensive than in normotensive persons.⁴ However, the opposite was found in a German population, where Arg16 was more common in hypertensive persons.⁵ Because of the problem of linkage disequilibrium in these polymorphisms, we studied β AR haplotypes in a population of white and blacks.⁶ We found no relationship between β_2 AR haplotypes and blood pressure in either population (Table I). Nor were there differences in allele frequencies between blacks and white Americans (Table II).

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Supported by grants HL-56521, HL-65962, GM-31304 from National Institutes of Health.

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0091-6749/2002 \$35.00 + 0 1/0/129701

doi:10.1067/mai.2002.129701

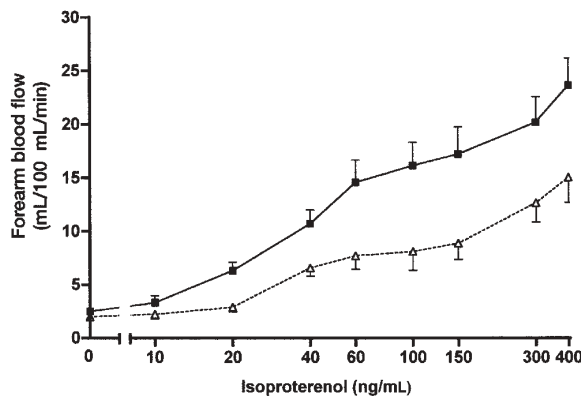


FIG 1. Forearm blood flow (mL/100 mL/min) at baseline and after the administration of increasing doses of intra-arterial isoproterenol (10-400 ng/mL) in normotensive controls (n = 12) and subjects with borderline hypertension (n = 8). Data are expressed as means $\pm$ SEM. ($P < .001$ [ANOVA]). Normotensive (■) and hypertensive (△). (From Stein CM, Nelson R, Deegan R, He H, Wood M, Wood AJJ. Forearm β -adrenergic receptor-mediated vasodilation is impaired, without alteration of forearm norepinephrine spillover, in borderline hypertension. J Clin Invest 1995;96:579-85. With permission.)

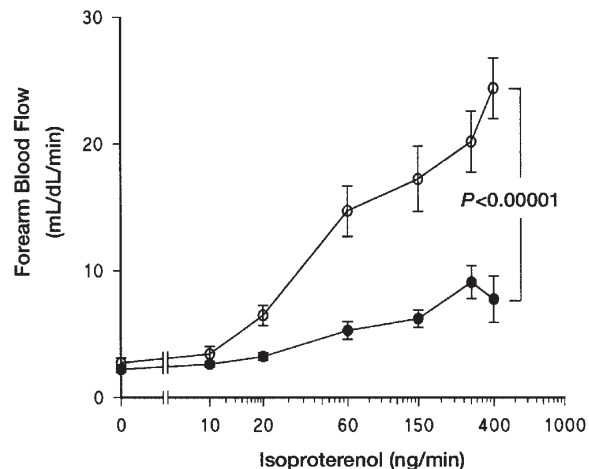


FIG 2. Forearm blood flow responses to isoproterenol administered by intrabrachial infusion in normotensive blacks (●) (n = 9) and whites (○) (n = 12). The rates of infusion ranged from 10 to 400 ng/min (log scale). The values shown are means $\pm$ SD. The increase in the forearm blood flow in response to isoproterenol was significantly blunted in blacks ($P < .001$). (From Lang CC, Stein CM, Brown RM, Deegan R, Nelson R, He HB, et al. Attenuation of isoproterenol-mediated vasodilation in blacks. N Engl J Med 1995;333:155-60. With permission.)

TABLE I. Frequencies of the β_2 AR combined genotypes in normotensive and hypertensive black and white American populations

Genotypes	Black				White			
	Normotensive (n = 128)		Hypertensive (n = 155)		Normotensive (n = 179)		Hypertensive (n = 201)	
Arg16/Arg16 + Gln27/Gln27	31	(24.2%)	48	(31.0%)	35	(19.6%)	45	(22.4%)
Arg16/Gly16 + Gln27/Gln27	39	(30.5%)	38	(24.5%)	25	(14.0%)*	27	(13.4%)
Arg16/Gly16 + Gln27/Glu27	18	(14.1%)	34	(21.9%)	56	(31.3%)*	59	(29.4%)
Gly16/Gly16 + Gln27/Gln27	18	(14.1%)	16	(10.3%)	18	(10.0%)	17	(8.4%)
Gly16/Gly16 + Gln27/Glu27	16	(12.5%)	15	(9.7%)	20	(11.2%)	21	(10.4%)
Gly16/Gly16 + Glu27/Glu27	6	(4.7%)	4	(2.6%)	25	(14.0%)	32	(15.9%)*

Data are expressed as n (%). There were no significant differences between normotensive and hypertensive groups ($P > .002$). Ethnic comparison (whites vs blacks): * $P < .001$. (From Xie HG, Stein CM, Kim RB, Gainer JV, Sofowora G, Dishy V, et al. Human β_2 -adrenergic receptor polymorphisms: no association with essential hypertension in black or white Americans. Clin Pharmacol Ther 2000;67:670-5. With permission.)

TABLE II. Frequencies of β_2 AR alleles and genotypes in hypertensive and normotensive black and white American populations

	Black		White	
	Normotensive (n = 128)	Hypertensive (n = 155)	Normotensive (n = 179)	Hypertensive (n = 201)
Allele				
Arg16	119 (46.5%)	168 (54.2%)	151 (42.2%)	176 (43.8%)
Gly16	137 (53.5%)	142 (45.8%)	207 (57.8%)	226 (56.2%)†
Gln27	210 (82.0%)	253 (81.6%)	232 (64.8%)	258 (64.2%)
Glu27	46 (18.0%)	57 (18.4%)	126 (35.2%)‡	144 (35.8%)‡
Genotype				
Arg16/Arg16	31 (24.2%)	48 (31.0%)	35 (19.6%)	45 (22.4%)
Arg16/Gly16	57 (44.5%)	72 (46.4%)	81 (45.2%)	86 (42.8%)
Gly16/Gly16	40 (31.3%)	35 (22.6%)	63 (35.2%)	70 (34.8%)*
Gln27/Gln27	88 (68.8%)	102 (65.8%)	78 (43.6%)‡	89 (44.3%)‡
Gln27/Glu27	34 (26.6%)	49 (31.6%)	76 (42.4%)†	80 (39.8%)
Glu27/Glu27	6 (4.7%)	4 (2.6%)	25 (14.0%)†	32 (15.9%)‡

Data are expressed as n (%). There were no significant differences between normotensive and hypertensive groups ($P > .05$). Ethnic comparison (whites vs blacks): * $P < .05$; † $P < .01$; ‡ $P < .0001$. (From Xie HG, Stein CM, Kim RB, Gainer JV, Sofowora G, Dishy V, et al. Human β_2 -adrenergic receptor polymorphisms: no association with essential hypertension in black or white Americans. Clin Pharmacol Ther 2000;67:670-5. With permission.)

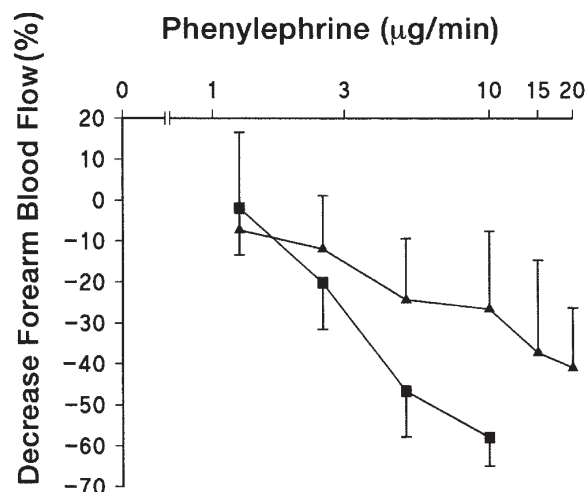


FIG 3. Percent decrease in forearm blood flow after intra-arterial infusion of 1.25 to 20 µg/min phenylephrine in normotensive blacks and whites. Values shown are mean $\pm$ SEM. Forearm blood flow response to phenylephrine was significantly increased in blacks (ANOVA, 12.25-10 µg/min phenylephrine) (race $\times$ dose, $P < .008$). Blacks (■) and whites (▲). (From Stein CM, Lang CC, Singh I, He HB, Wood AJJ. Increased vascular adrenergic vasoconstriction and decreased vasodilation in Blacks: additive mechanisms leading to enhanced vascular reactivity. Hypertension 2000;36:945-51. With permission.)

TABLE III. Effect of genotype on vasodilatory response to isoproterenol before desensitization

Variable	Homozygous for Arg16 and Gln27 (n = 7)	Homozygous for Gly16 and Gln27 (n = 8)	Homozygous for Gly16 and Glu27 (n = 11)
Maximal vasodilative response to isoproterenol (%)*	50 $\pm$ 11	57 $\pm$ 11	86 $\pm$ 13‡
Dose of isoproterenol producing 20% venodilation (ng/min)†	43 (7-276)	62 (25-158.5)	16 (6-40)
Dose of isoproterenol producing maximal venodilation (ng/min)†	81 (19-350)	195 (132-288)	170 (94-302)

*Percentage reversal of phenylephrine-induced vasoconstriction, expressed as means $\pm$ SE.

†Expressed as geometric means (95% confidence intervals).

‡ $P = .03$ for the comparison of subjects who were homozygous for Glu27 with those who were homozygous for Gln27 (maximal vasodilative response, 54% $\pm$ 8%).

(From Dishy V, Sofowora GG, Xie H-G, Kim RB, Byrne DW, Stein CM, et al. The effect of common polymorphisms of the β_2 -adrenergic receptor on agonist-mediated vascular desensitization. N Engl J Med 2001;345:1030-5. With permission.)

However, marked ethnic differences were seen in the frequencies of the different polymorphisms (Tables I and II). We questioned whether these inter-ethnic differences in genotype frequency might offer a partial explanation for the impaired vasodilatation in blacks.²

Because the polymorphisms were thought to be associated with altered ability of the receptor to desensitize in response to agonist exposure, we studied groups of per-

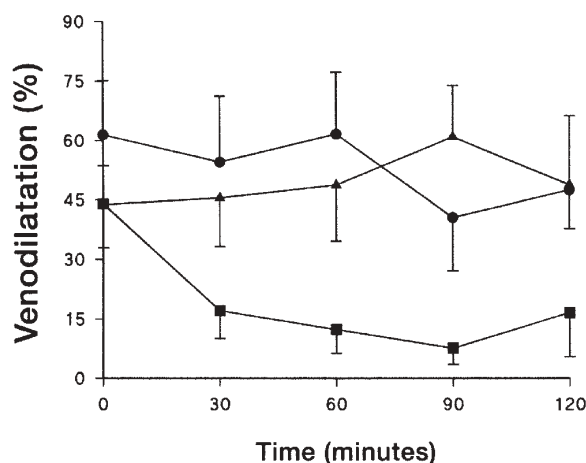


FIG 4. Mean $\pm$ SEM venodilation during continuous infusion of isoproterenol (desensitization) in the three groups of subjects. There was a significant interaction between genotype and the response to continuous infusion of isoproterenol ($P = .03$ by repeated-measures ANOVA). Subjects who were homozygous for Arg16 and Gln27 had significant desensitization ($P = .006$), whereas subjects who were homozygous for Gly16 and Gln27 and those who were homozygous for Gly16 and Glu27 did not have desensitization over time ($P = .56$ and $P = .11$, respectively). (●) Homozygous for Gly16 and Glu27, (▲) homozygous for Gly16 and Gln27, (■) homozygous for Arg16 and Gln27. (From Dishy V, Sofowora GG, Xie H-G, Kim RB, Byrne DW, Stein CM, et al. The effect of common polymorphisms of the β_2 -adrenergic receptor on agonist-mediated vascular desensitization. N Engl J Med 2001;345:1030-5. With permission.)

sons matched for haplotypes and determined the response of their hand vein to chronic exposure to isoproterenol.⁷ Three haplotypes were studied. Seven persons were homozygous for Arg16 and Gln27, 8 were homozygous for Gly16 and Gln27, and 11 were homozygous for Gly16 and Glu27. Almost complete desensitization occurred in persons who were homozygous for Arg16. Irrespective of the presence of Gln or Glu at position 27, persons who were homozygous for Gly16 had no significant desensitization in response to chronic agonist administration (Fig 4).⁷ Persons who were homozygous for Glu27 had a greater maximal vasodilatory response to isoproterenol than those who were homozygous for Gln27 (Table III). This is of great interest as this homozygosity is less prevalent in blacks than in white Americans, providing a potential explanation for the impaired vasodilatation seen in blacks.

CONCLUSIONS

It appears that β AR polymorphism may be an important determinant of vascular response in humans. Further studies will be required to determine the contribution of these polymorphisms to vascular disease.

DISCUSSION SESSION

Question: Can you reconcile your in vivo data with what is in the in vitro literature regarding the Arg 16/Gly 16 polymorphism?

Dr Wood: One potential explanation is that the two haplotypes that do not desensitize are in fact the most sensitive haplotypes. They already are desensitized to such a degree that they cannot be desensitized further. The people who are the least sensitive to desensitization are those in whom we see the desensitization. The tissues we are studying—the vasculature—are bathed in catecholamines. So, it is very important to control for catecholamine concentrations. We do this by placing everybody on a constant sodium intake. We have used this approach to manipulate catecholamine concentrations chronically. If you put a patient on a low-sodium diet, the catecholamines increase substantially. That is how the patient maintains vasoconstriction and does not experience a substantial drop in blood pressure. Conversely, if you put a patient on a high-salt diet, the catecholamines decrease. One can manipulate people over a physiological range of catecholamines by using that technique. We have used this approach to uncouple and desensitize receptors. So, we could go back and, before giving isoproterenol, change catecholamine concentrations by subjecting patients to very high or very low catecholamine concentrations.

Question: Would it be useful to study receptor gene variations in people who have the most difficulty with hypoglycemic unawareness?

Dr Wood: It would be interesting to assess the polymorphisms and see if the desensitization to β -agonists that occurs differs.

Question: You use isoproterenol as your dilator and phenylephrine as your constrictor. Phenylephrine is purely α_1 , whereas isoproterenol is β_1 and β_2 . Are there any important post-synaptic differences between blacks and whites, for example, that are α_2 -based, either in the vessel itself or in the central nervous system?

Dr Wood: Yes. We have given α_2 -agonists to blacks and whites and shown that the effects differ quite substantially. The difficulty in interpreting that experiment is that when you give α_2 -agonists systemically, you lower catecholamine concentrations. The vasoconstrictive effects of α_2 -agonists are clear. When we have constructed dose response curves to intravenous clonidine, for example, we observe that blacks have a different, catecholamine-dependent response.

Question: Why do patients with hypertension have an altered vascular response to constrictors and dilators?

Dr Wood: I am uncertain. We have studied many vasodilators and vasoconstrictors, and it is not a uniform effect. It appears that there is a difference in the response to a number of different agonists. There is no explanation for this yet.

Question: Are there any clinical features of the hypertensive asthmatic or the hypertensive COPD patient that distinguish them from patients with airways disease who are not hypertensive? Are hypertensives with asthma more difficult to care for than hypertensives without asthma?

Dr Wood: At a practical level they are because we cannot use vasodilators easily, and we cannot give these patients β blockers. We use α_2 -agonists because of the unwillingness to give β blockade.

Question: Do asthmatics have altered forearm blood flow responses to these agents?

Dr Wood: Yes, that would be an easy test. It could even be measured in the hand vein. In fairness, we should not assume that all

vascular beds are identical or that the hand vein is the same as the brachial artery. The vessels that determine blood pressure in terms of being responsible for the peripheral vascular resistance may be different from the bronchi that determine airway resistance. In heart failure, for example, the changes in the veins may be different from changes in the artery, which may be different from the small arteries.

Question: Have you studied the microcirculation to assess the effects on vascular permeability in response to β -agonists?

Dr Wood: We have not studied this. The brachial artery infusions are very dependent on microvascular changes. We are not looking directly at the brachial artery diameter, but at forearm blood flow, which is determined by resistance. In the hand vein we are measuring the effect within less than a centimeter from the drug infusion site. People have used skin temperature as a measure of small vessel changes. This is not a very sensitive measure.

Question: Some of the drugs that are used to treat hypertension act at the central nervous system level. Are there apparent differences or reasons why some people are more responsive in this way or is it just part of the syndrome of hypertension?

Dr Wood: Well, some of the α_2 effects of clonidine are centrally mediated. People used to believe that bad thoughts caused hypertension. However, this is not the case. Many patients with hypertension do not seem to lead a very high-stress life.

Question: Does pruning of the small blood vessels distal to the brachial artery occur in hypertensives? Is it variable enough to influence the results and their interpretation?

Dr Wood: In severe hypertension, pruning of the arteries does occur. This is classically seen in the kidneys. However, at the mild level of hypertension studied here, pruning is not seen.

REFERENCES

1. Stein CM, Nelson R, Deegan R, He H, Wood M, Wood AJJ. Forearm beta adrenergic receptor-mediated vasodilation is impaired, without alteration of forearm norepinephrine spillover, in borderline hypertension. *J Clin Invest* 1995;96:579-85.
2. Lang CC, Stein CM, Brown RM, Deegan R, Nelson R, He HB, et al. Attenuation of isoproterenol-mediated vasodilatation in blacks. *N Engl J Med* 1995;333:155-60.
3. Stein CM, Lang CC, Singh I, He HB, Wood AJJ. Increased vascular adrenergic vasoconstriction and decreased vasodilation in blacks: additive mechanisms leading to enhanced vascular reactivity. *Hypertension* 2000;36:945-51.
4. Kotanko P, Binder A, Tasker J, DeFreitas P, Kamdar S, Clark AJL, et al. Essential hypertension in African Caribbeans associates with a variant of the β_2 -adrenoceptor. *Hypertension* 1997;30:773-6.
5. Timmermann B, Mo R, Luft FC, Gerdts E, Busjahn A, Omvik P, et al. β -2 adrenoceptor genetic variation is associated with genetic predisposition to essential hypertension: the Bergen Blood Pressure Study. *Kidney Int* 1998;53:1455-60.
6. Xie HG, Stein CM, Kim RB, Gainer JV, Sofowora G, Dishy V, et al. Human β_2 -adrenergic receptor polymorphisms: no association with essential hypertension in black or white Americans. *Clin Pharmacol Ther* 2000;67:670-5.
7. Dishy V, Sofowora GG, Xie H-G, Kim RB, Byrne DW, Stein CM, et al. The effect of common polymorphisms of the β_2 -adrenergic receptor on agonist-mediated vascular desensitization. *N Engl J Med* 2001;345:1030-5.