

The interaction of nitric oxide, bradykinin, and the angiotensin II type 2 receptor: lessons learned from transgenic mice

Commentary

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Despite intensive study over the past few years, several aspects of the renin-angiotensin system remain poorly understood. Such is the case for the angiotensin II type 2 receptor (AT₂), one of the two receptor subtypes that mediate the actions of angiotensin II. In fetal tissues, this receptor is expressed at high levels and appears to have a role in growth, differentiation, and maturation of cells in various organs, including the vasculature (1–4). As development progresses, the level of expression of the AT₂ is downregulated. In adult animals, in contrast to the wide distribution of angiotensin II type 1 receptor (AT₁), AT₂ is expressed at low levels and is restricted to the adrenal gland, brain, ovary, uterus, kidney, and heart. Importantly, there appears to be upregulation of AT₂ in pathological states such as salt depletion, heart failure, experimental cardiac hypertrophy, myocardial infarction, and vascular injury (3–8).

Because of the low level of AT₂ expression in normal tissues, there is substantial debate as to its role under normal circumstances. A growing body of research suggests that there is crosstalk between AT₁ and AT₂ in mediating the physiologic effects of angiotensin II. Based largely on pharmacological studies, stimulation of the AT₂ seems to antagonize several of the effects caused by AT₁ stimulation. Thus, AT₁ stimulation activates the mitogen-activated protein kinases ERK1 and ERK2, whereas AT₂ stimulation suppresses this pathway, perhaps by activating ERK phosphatase 1 (MKP-1) (4, 9, 10). AT₁ stimulation promotes cellular growth and hypertrophy, while the AT₂ antagonizes them (3–6). AT₁ stimulation facilitates angiogenesis, while the AT₂ inhibits this process (11). AT₁ activation induces vasoconstriction, while AT₂

activation causes vasodilation (12). These cellular and organ-level effects appear to act in intact animal models as well. In cardiomyopathic hamsters, AT₂ expression is upregulated in cardiac fibroblasts of the failing heart and appears to antagonize AT₁-mediated progression of interstitial fibrosis and cardiac remodeling (6). Overexpression of AT₂ in balloon-injured vascular smooth muscle cells attenuates neointimal formation (6). In a rat model of ischemic cardiomyopathy, the beneficial effects of AT₁ blockade on cardiac remodeling and hemodynamics are inhibited by AT₂ blockade (5).

Recent work has suggested that some of the beneficial effects of AT₂ stimulation may be mediated through the bradykinin/nitric oxide (NO) cascade (5, 13–15). Endothelial cells contain bradykinin type 2 receptors (B₂), which, when activated, potently stimulate production of NO. Although the effects of angiotensin II and NO seem to vary with the concentration of NO and the cell type involved, these two factors appear to play opposing roles in the cardiovascular system: angiotensin II is a potent stimulus for vasoconstriction and vascular smooth muscle hypertrophy, whereas NO has a vasodepressor effect and has been shown to be an antiproliferative agent. Thus, in spontaneously hypertensive rats, AT₂ activation has been shown to increase vascular cyclic guanosine 3',5'-monophosphate (cGMP) levels, an effect that could be inhibited by B₂ blockade or by inhibition of NO synthase (15). Likewise in conscious rats, salt depletion, which activates the renin-angiotensin system, increases cGMP levels in the renal interstitial fluid (14), an effect that can be prevented by blockade of either NO synthase or the AT₂. Following myocardial infarction in rats, either

angiotensin I-converting enzyme inhibitors or AT₁ antagonists can prevent remodeling of the left ventricle, as assessed by collagen deposition, myocyte size, and left ventricular diameter (5), and in either case the effect could be blocked by B₂ inhibition. Importantly, these studies not only underscore the antagonistic interactions between AT₁ and AT₂ but also introduce the notion that the bradykinin/NO system mediates this interaction.

In the previous issue of the *JCI*, Tsutsumi et al. (16) provided additional compelling evidence linking AT₂ to the bradykinin/NO cascade. These authors targeted overexpression of AT₂ to the vascular smooth muscle in transgenic mice, achieving a 5-fold increase in expression of this receptor. These animals exhibited an attenuated pressor response to angiotensin II infusion. Pretreatment of these transgenic mice with an AT₂ antagonist, a B₂-receptor antagonist, or an NO synthase inhibitor restored the pressor response to angiotensin II. Angiotensin II produced a paradoxical decrease in blood pressure after AT₁ blockade in these animals, suggesting that selective AT₂ stimulation had a vasodepressor effect. Furthermore, the authors showed that the AT₂-mediated vasodepressor effect was associated with an endothelium-dependent increase in aortic production of cGMP and activation of the kinin-kallikrein system. In the authors' interpretation of the results, angiotensin II stimulates AT₂ in vascular smooth muscle, which leads to activation of the kinin-kallikrein system and bradykinin release. Bradykinin then binds to its receptor on adjacent endothelial cells, causing the release of NO and stimulation of cGMP.

As with other work investigating the physiologic role of AT₂, this study

relied heavily on pharmacologic manipulation, and the question arises as to what extent the results could be attributed to the specificity of the various agents used. Despite this concern, the transgene clearly altered the effects of exogenously administered angiotensin II, and therefore this work provides important information regarding interactions between angiotensin II and NO in the vasculature. This study is the first to directly demonstrate AT2 stimulation of vascular smooth muscle kininogenase activity, which, in turn, explains increased vascular production of bradykinin and NO in response to angiotensin II. Certainly, more work has to be done to verify and elucidate the physiologic importance of this angiotensin II/bradykinin/NO cascade and its relevance to heart failure, myocardial infarction, and vascular injury.

One intriguing notion that has evolved from this type of work is that failure of normal crosstalk between AT1 and AT2 may worsen development of cardiovascular disease. Thus, absence or decreased activity of AT2 may allow the deleterious effects of AT1 stimulation to go unchecked. Interestingly, NO has been reported to downregulate AT1 expression (17). Therefore, enhanced NO production in response to AT2 stimulation may diminish AT1 respon-

siveness directly. Conversely, selective pharmacotherapeutic stimulation of the AT2 may have beneficial effects in treatment of cardiovascular diseases. Treatment with AT1 antagonists, which leave AT2 unblocked, may in part achieve this end. As the role of the AT2 rises from physiologic obscurity, a more thorough understanding of its interactions with the AT1 and the NO system is emerging. Understanding these interactions may allow greater insight into the full impact of angiotensin II in disease.

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