Letters to the Editor

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No Net Effect of Angiotensin II on Blood Pressure?

To the Editor:

With great interest we read the recent study by Carey et al[1] on angiotensin (Ang) II type 2 receptor (AT_2) receptor–mediated hypotension in anesthetized and conscious rats. These data oppose our findings on the lack of AT_2 receptor–mediated systemic hemodynamic effects in anesthetized rats.[2] To explain this discrepancy, Carey et al propose that the doses of the Ang II type 1 (AT_1) and AT_2 receptor antagonists used in our study (irbesartan and PD123319, respectively) were insufficient to unmask AT_2 receptor–mediated vasodilation. However, the same doses were high enough to observe AT_2 receptor–mediated vasodilation in the rat coronary vascular bed.[3] Furthermore, the dose of irbesartan that we used was high enough to fully block the Ang II–induced vasoconstrictor effects in our study, as well as in studies by others.[4] Moreover, the PD123319 dose that we used is known to result in micromolar concentrations in blood plasma,[5] ie, concentrations that are high enough to selectively block AT_2 receptors. Higher doses lead to concentrations that also interfere with AT_1 receptors.[5]

Carey et al mention that AT_2 receptors in the adult rat are present in low copy compared with AT_1 receptors. In agreement with this concept, exogenous Ang II induces vasoconstriction in the absence of AT receptor blockers, and AT_1 receptor blockade with either losartan or valsartan results in vasodilation. Unfortunately, the authors did not study the effect of PD123319 alone. If, indeed, Ang II normally has a net vasoconstrictor effect, based on the much larger density of AT_1 receptors compared with AT_2 receptors, full blockade of both AT_1 and AT_2 receptors should result in a net *decrease* in blood pressure. Remarkably, however, the authors observe a normal blood pressure during combined AT_1 and AT_2 receptor blockade, as if normally Ang II has no net effect on blood pressure.

In this regard, it is important to realize that combined infusions of AT receptor antagonists have been reported to interfere with the effective plasma and tissue levels of the drugs[6] and that PD123319 is transformed to a metabolite with AT_1 receptor antagonist properties in vivo.[7] Thus, pharmacokinetic interactions should be considered when interpreting the data by Carey et al, particularly because these studies lasted several days (thus allowing significant generation of interfering metabolites), and the AT_2 receptor–mediated effects in conscious animals became maximal after only 8 days.

Finally, the systemic vasodilation observed by Carey et al with the partial AT_2 receptor agonist CGP-42112 is in full agreement with the CGP-42112–induced renal vasodilator responses that were described earlier by Macari et al.[5] In the latter study, however, this effect was ascribed to the AT_1 receptor–blocking capacity of CGP-42112. The nitric oxide (NO) synthase inhibitor, N^G-nitro-L-arginine methylester (L-NAME), reversed the CGP-42112–induced vasodilation, leading Carey et al to suggest that the AT_2 receptor–mediated effects depend on NO. However, physiological receptor antagonism, ie, 2 independent opposite effects, cannot be excluded as an explanation for this finding. Furthermore, it is difficult to understand why the authors, in contrast to many previous studies, did not observe a hypertensive effect of L-NAME.

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Response

We appreciate the interest of Schuijt et al in our paper, "Angiotensin II type 2 receptor–mediated hypotension in angiotensin type-1 receptor–blocked rats."[1] As we stated in our report, several studies have failed to demonstrate AT_2 receptor–mediated reduction in blood pressure (BP) in the intact rat. Our explanation for this discrepancy was that these previous studies most likely used much smaller doses of the AT_2 receptor blocker, PD 123319, than our study and never demonstrated that PD 123319 fully blocked the AT_2 receptor. In our study we used PD 123319 at 50 μg/kg per minute, an infusion rate that blocked specifically the AT_2 receptor as demonstrated by its ability to decrease renal production of nitric oxide and cGMP without influencing AT_1 receptor activity.[2,3] The affinity of PD 123319 for the AT_2 receptor is about 17 nmol/L.[4–7] Previously, we demonstrated that the AT_1 receptor is responsible for prostaglandin E_2 (PGE_2) release in the kidney[1] and that PD 123319 50 μg/kg per minute did not reduce renal PGE_2.

Schuijt et al correctly observed that we did not study the effect of PD 123319 alone on BP. However, it had already been demonstrated that PD 123319 enhances the pressor effect of Ang II.[8] Previous studies also demonstrated that animals lacking the AT_2 receptor exhibit higher BP than their wild-type controls.[9–12] Schuijt et al state that PD 123319 is transformed to a metabolite with AT_1 receptor antagonist properties. This could happen only

if PD 123319 were used in heroic doses that approach the Ki for the AT_1 receptor at about 100 μmol.[13] In our study, the PD 123319 infusion rate was <1 μmol/kg per minute, a concentration far below the Ki for the AT_1 receptor.

The concept that full blockade of both AT_1 and AT_2 receptors should result in a net decrease in BP is flawed, because several other compensatory mechanisms would be expected to maintain normal BP.

Concerning the effects of the AT_2 receptor agonist CGP-42112 (CGP), it is known that CGP at large doses (>1000 μg/kg per minute) can cross over to influence the AT_1 receptor. Previous studies reported that both CGP and PD 123319 have a similar Ki for the AT_2 receptor.[14] Macari et al reported that large doses of CGP block[14] and stimulate[15] the AT_1 receptor. Thus, it is very important to limit the CGP dose to less than 1 μmol/L. In our studies the CGP dose was confined within this limitation. Recent studies by Bautista et al[16] confirm this conclusion and clearly show that CGP at 1 μmol/L has renal vasodilator effects through stimulation of the AT_2 receptor.

The AT_2 receptor has a very high affinity for CGP with IC_{50} of $4+2.2$ nmol, whereas AT_1 receptor has a low affinity for CGP with an IC_{50} of $11.4+1.9$ μmol.[17] An approximately 3000-fold difference in affinity exists between the AT_2 and AT_1 receptors for the AT_2-selective agonist CGP. High micromolar concentrations of CGP that completely displace Ang II from AT_1 receptor in response to CGP is not an AT_1-receptor attribute, but rather should be considered an AT_2-receptor –specific phenomenon.[17] All these data confirm the specificity and effects of the AT_2 receptor agonist CGP at the infusion rates employed in our study.

Schuijt et al question why N^G-nitro-L-arginine methylester (L-NAME) did not produce a hypertensive effect. We did not administer L-NAME alone in our study. However, L-NAME did increase BP during CGP administration.

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