Angiotensin II Enhances Connecting Tubule Glomerular Feedback

YiLin Ren, Martin A. D’Ambrosio, Jeffrey L. Garvin, Oscar A. Carretero

Abstract—Increasing Na delivery to epithelial Na channels (ENaCs) in the connecting tubule (CNT) causes dilation of the afferent arteriole (Af-Art), a process we call CNT glomerular feedback (CTGF). Angiotensin II (Ang II) stimulates ENaC in the collecting duct via Ang II type 1 receptors. We hypothesized that Ang II in the CNT lumen enhances CTGF by activation of Ang II type 1 receptors, protein kinase C and ENaC. Rabbit afferent arterioles and their adherent CNT were microperfused and preconstricted with norepinephrine. Each experiment involved generating 2 consecutive concentration-response curves by increasing NaCl in the CNT lumen. During the control period, the maximum dilation of the afferent arteriole was 7.9±0.4 μm, and the concentration of NaCl in the CNT needed to achieve half maximal response (EC50) was 34.7±5.2 mmol/L. After adding Ang II (10−9 mol/L) to the CNT lumen, the maximal response was 9.5±0.7 μm and the EC50 was 11.6±1.3 mmol/L (P=0.01 versus control). Losartan, an Ang II type 1 antagonist (10−6 mol/L) blocked the stimulatory effect of Ang II; PD123319, an Ang II type 2 antagonist (10−6 mol/L), did not. The protein kinase C inhibitor staurosporine (10−8 mol/L) added to the CNT inhibited the stimulatory effect of Ang II. The ENaC inhibitor benzamil (10−6 mol/L) prevented both CTGF and its stimulation by Ang II. We concluded that Ang II in the CNT lumen enhances CTGF via activation of Ang II type 1 and that this effect requires activation of protein kinase C and ENaC. Potentiation of CTGF by Ang II could help preserve glomerular filtration rate in the presence of renal vasoconstriction. (Hypertension. 2010;56:636-642.)

Key Words: angiotensin II ▪ distal kidney tubules ▪ arterioles ▪ renin-angiotensin system ▪ angiotensin II type 1 receptor blockers

At least 2 segments of the distal nephron return to their original glomerulus, where they come in contact with the afferent arteriole (Af-Art). The macula densa contacts the vascular pole of the glomerulus, where it is well established that it controls glomerular hemodynamics via tubuloglomerular feedback (TGF).1,2 In most nephrons, a later segment of the distal nephron, the connecting tubule (CNT), also contacts the Af-Art of the same nephron.3-6 We have recently described the existence of cross-talk between the CNT and the Af-Art, which we called CNT glomerular feedback (CTGF).7,8 Both TGF and CTGF are initiated by increases in Na concentration in the tubular lumen, but TGF causes vasoconstriction and CTGF vasodilation. In the presence of an increased sodium load in the distal nephron, such as volume expansion or high-salt intake, activation of TGF causes Af-Art constriction, which tends to hinder sodium excretion by decreasing glomerular filtration rate (GFR). However, additional mechanisms come into play, whereby in volume expansion TGF is desensitized, favoring sodium excretion.9 Such resetting mechanisms are not entirely understood but may include the newly described CTGF. CTGF has an opposite effect to TGF in that in the presence of high distal sodium it tends to dilate the Af-Art.

In the kidney, a local tubular renin angiotensin system has been described. Angiotensinogen secreted into the lumen of the proximal tubule can reach the distal nephron.10 Renin expressed in the CNT and collecting duct11 can then convert angiotensinogen to angiotensin I. Angiotensin converting enzyme, present in the tubular fluid can generate angiotensin II (Ang II).12 Ang II acts via 2 different receptors, Ang II type 1 (AT1) and Ang II type 2 (AT2), both types are expressed in the CNT and collecting duct11 can then convert angiotensinogen to angiotensin I. Angiotensin converting enzyme, present in the tubular fluid can generate angiotensin II (Ang II). Ang II can also potentiate CTGF via activation of Ang II type 1 and that this effect requires activation of protein kinase C and ENaC. Potentiation of CTGF by Ang II could help preserve glomerular filtration rate in the presence of renal vasoconstriction. (Hypertension. 2010;56:636-642.)

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in collecting ducts, and we have shown that Na entry via ENaC in the CNT initiates CTGF. 

The signaling pathway(s) activated by Ang II in the CNT remains unclear, but in other nephron segments Ang II activates the phospholipase C signaling pathway, thereby elevating cytosolic Ca²⁺ and protein kinase C (PKC). PKC inhibitors suppress Ang II–induced Na⁺ transport and fluid reabsorption in proximal tubular cells.

In the present study, we hypothesize that Ang II in the CNT lumen enhances CTGF via stimulation of AT1 receptors, which require activation of PKC, and ENaC. To test this hypothesis while avoiding the confounding influence of the multiple systemic factors that regulate the renal microcirculation, we simultaneously perfused a microdissected rabbit Af-Art and adherent CNT.

**Methods**

New Zealand White rabbits weighing 1.5 to 2.0 kg (Myrtle’s Rabbity, Thompson’s Station, TN) were given standard chow (Ralston Purina) and tap water ad libitum and anesthetized with ketamine (50 mg/kg IM), xylazine (10 mg/kg IM), and pentobarbital (25 mg/kg IV). All of the protocols were approved by the Henry Ford Health System Institutional Animal Care and Use Committee and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals. We used rabbits because their CNTs are well demarcated, and microdissection of the CNT and attached Af-Art is easier than in rats or mice. To isolate and microperfuse the Af-Art and CNT, we used methods similar to those described previously. This system allows us to exchange the perfusion solution in a few seconds while keeping the holding and perfusion pipettes in place. The Af-Art was perfused with minimum essential medium containing 5% BSA (Sigma). Using fine forceps, a single superficial Af-Art with its glomerulus intact was dissected together with the adherent CNT. Using a micropipette, the microdissected complex was transferred to a temperature-regulated perfusion chamber mounted on an inverted microscope with Hoffmann modulation. Both the Af-Art and CNT were cannulated with an array of concentric glass pipettes, as described previously. This system allows us to exchange the perfusion solution in a few seconds while keeping the holding and perfusion pipettes in place. The Af-Art was perfused with minimum essential medium containing 5% BSA gassed with room air. Intraluminal pressure was measured by the Landis technique and maintained at 60 mm Hg. The CNT perfusion solution contained (in millimoles per liter): 4 KHCO₃, 10.0 HEPES, 0.5 Na acetate, 0.5 Na lactate, 0.5 K₂HPO₄, 1.2 MgSO₄, 1.0 CaCl₂, and 5.5 glucose, adding 1 mol/L of NaCl to achieve the desired final NaCl concentration. Tubular perfusion was controlled by use of a syringe microperfusion pump (Harvard Apparatus Inc) set to 20 nL/min (calibration checked to be 10–20 nL/min), which is within the range of physiological flow rates. The bath was superfused with minimum essential medium containing 0.15% BSA at a rate of 1 mL/min.

Microdissection and cannulation of the Af-Art and CNT were completed within 90 minutes at 8°C, after which the temperature was gradually raised to 37°C. Once it was stable, a 30-minute equilibration period was allowed before taking any measurements. Because isolated arteries have little or no tone and our preliminary studies showed that increasing NaCl in the CNT perfusate caused only modest dilation of the Af-Art unless it was preconstricted, we performed all of the CTGF experiments with Af-Arts preconstricted with norepinephrine (NE; 2 to 5 × 10⁻⁷ mol/L). In each experiment 2 consecutive concentration-response curves were generated by increasing luminal NaCl in the CNT from 0 to 5, 10, 30, 45, and 80 mmol/L. Af-Art diameter was measured in the region of maximal vasoactivity at 20 mmol/L of NaCl, which is within the range of physiological flow rates. The bath caused a dose-dependent vasoconstriction of the Af-Art, unless it was preconstricted, we carried out some experiments with Af-Arts left unprecontracted, to provide further evidence that Ang II potentiates CTGF.

To demonstrate that the CTGF response is stable over time, we performed 2 consecutive concentration-response curves to Ang II (10⁻⁹ to 10⁻⁵ mol/L) in the presence of different Ang II concentrations. The Af-Art was perfused with 0 NaCl, Ang II added to the CNT did not affect Af-Art diameter. However, when CNT NaCl was increased in the presence of Ang II, CTGF-induced dilation of the Af-Art was potentiated, that is, the curve was shifted to the left (maximal response: 9.5±0.7 μm; EC₅₀: 11.6±1.3 mmol/L; P<0.01 versus control; Figure 1B). These data demonstrate that intratubular Ang II enhances the CTGF response.

To determine whether increasing Ang II in the CNT causes CTGF during constant NaCl delivery, we performed a dose-response curve to Ang II (10⁻¹² to 10⁻⁹ mol/L) perfusion in the CNT, while the concentration of NaCl was maintained constant at 20 mmol/L of NaCl, that is, a NaCl concentration that does not induce CTGF, per se. The addition of Ang II induced CTGF in a dose-dependent manner; perfusion of the CNT with Ang II 10⁻¹⁰ mol/L tended to potentiate CTGF, with further potentiation seen with Ang II 10⁻⁹ mol/L (Figure 1C). These data provide further evidence that Ang II potentiates CTGF.

Values are expressed as mean±SEM. Paired t tests were used to compare CTGF (ΔAf-Art diameter) between the control and experimental periods. The Hochberg step-up procedure was used to adjust the P values for multiple comparisons. EC₅₀ calculation was performed by best-fit regression analysis and 4 parameter model (XLSTAT software).

**Results**

To show that the CTGF response remains stable over time, we performed 2 consecutive concentration-response curves to 0 to 5, 10, 30, 45, and 80 mmol/L of NaCl in the lumen of the CNT. As shown in Figure 1A, NE caused a 45% decrease in Af-Art diameter when the attached CNT was perfused with 0 sodium. Increasing NaCl concentration in the CNT relaxed the Af-Art in a concentration-dependent manner, restoring basal diameter at 80 mmol/L of NaCl. The maximal vasodilatory response, measured as the increase in diameter of preconstricted Af-Arts in response to NaCl perfusion of the attached CNT (maximal response), was 8.1±0.6 μm. The concentration of NaCl in the CNT perfusate needed to achieve half of the maximal response (EC₅₀) was 41.1±1.6 mmol/L. We then returned CNT luminal NaCl to 0 and repeated the stepwise increase in NaCl concentration; the maximal response was 8.7±0.6 μm, and the EC₅₀ was 42.4±0.9 mmol/L. Thus, the 2 curves were similar, confirming that the CTGF response is stable over time.

To study the effect of intratubular Ang II on CTGF, we repeated the above-described experiment but added 10⁻⁹ mol/L of Ang II to the CNT perfusate during the second period. During the control period, increasing NaCl dilated the Af-Art in a concentration-dependent manner (maximal response: 7.9±0.4 μm; EC₅₀: 34.7±5.2 mmol/L). When the CNT was perfused with 0 NaCl, Ang II added to the CNT did not affect Af-Art diameter. However, when CNT NaCl was increased in the presence of Ang II, CTGF-induced dilation of the Af-Art was potentiated, that is, the curve was shifted to the left (maximal response: 9.5±0.7 μm; EC₅₀: 11.6±1.3 mmol/L; P<0.01 versus control; Figure 1B). These data demonstrate that intratubular Ang II enhances the CTGF response.
80 mmol/L and repeated the dose-response curve to Ang II in the bath, vasoconstriction was blunted. The maximum decrease in Af-Art diameter was 15% (from 16.9±0.9 to 14.3±1.2 µm; Figure 1D). These data indicate that CTGF is not only able to counter NE-induced but also Ang II-induced constriction of AF-Arts.

Because the effects of Ang II are mediated by AT₁ or AT₂ receptors, we questioned which one contributes to its effects on CTGF. We first compared Ang II alone with Ang II plus the AT₁ blocker losartan. With Ang II alone, increasing NaCl dilated preconstricted Af-Arts (maximal response: 10.7±1.6 µm; EC₅₀: 14.3±1.6 mmol/L). Adding losartan (10⁻⁶mol/L) to the CNT perfusate attenuated NaCl-induced dilation and the curve shifted to the right (maximal response: 8.6±1.3 µm; EC₅₀: 37.1±1.9 mmol/L; *P<0.001 versus Ang II alone; Figure 2A). These data show that AT₁ receptors mediate, at least in part, Ang II–induced potentiation of CTGF. To study whether blocking AT₁ receptors could

Figure 1. A, Increasing NaCl concentration in the CNT dilated preconstricted Af-Arts in a similar manner, indicating that CTGF is stable and reproducible over time. B, Adding 10⁻⁹ mol/L of Ang II to the CNT perfusate enhanced CTGF (**P<0.05; ***P<0.01, with vs without Ang II). C, In the presence of a low NaCl concentration, which does not elicit CTGF per se, increasing CNT Ang II concentrations dose-dependently dilated preconstricted Af-Art (**P<0.01, 10⁻⁹ M Ang II versus without Ang II). D, Adding 80 mmol/L of NaCl to the CNT perfusate blunted the constrictor response to Ang II applied to the attached Af-Art, suggesting that CTGF opposes circulating vasoconstrictors (**P<0.01, 10 mmol/L vs 80 mmol/L of NaCl).

Figure 2. A, Adding 10⁻⁶ mol/L of losartan to the CNT perfusate blocked Ang II–enhanced CTGF, suggesting that the effect of Ang II on CTGF is mediated by the AT₁ receptor. (**P<0.01; ***P<0.001, with vs without losartan). B, In the presence of Ang II plus losartan, CTGF did not differ from control, suggesting that the effect of Ang II on CTGF is mediated solely by the AT₁ receptors. C, Losartan alone did not affect CTGF in the absence of exogenous Ang II. D, Adding 10⁻⁶ mol/L of PD123319 (PD) to the CNT perfusate did not block Ang II–enhanced CTGF, suggesting that the effect of Ang II on CTGF is not mediated by the AT₂ receptors.
completely abolish the effect of Ang II, we first performed a control CTGF response, followed by coadministration of Ang II and losartan during the second period. We found that losartan completely prevented Ang II–induced enhancement of CTGF. During the control period the maximal response was 7.9 ± 1.2 μm and the EC_{50} was 30.9 ± 1.8 mmol/L. During Ang II plus losartan, the maximal response was 7.8 ± 0.9 μm and the EC_{50} was 31.5 ± 1.2 mmol/L (Figure 2B). In control experiments, we tested the effect of losartan on CTGF in the absence of exogenous Ang II. During the control period the maximal response was 7.4 ± 1.5 μm and the EC_{50} was 41.0 ± 1.4 mmol/L. During losartan administration, the maximal response was 7.5 ± 1.4 μm and the EC_{50} was 39.8 ± 0.6 mmol/L (Figure 2C). These data show that losartan does not affect CTGF in the absence of exogenous Ang II; it also suggests that endogenous Ang II is absent or minimal in our isolated preparation.

The fact that losartan completely blocked Ang II–induced potentiation of CTGF suggests that this effect is solely mediated by AT_{1} receptors. To confirm that CNT AT_{2} receptors are not involved in CTGF, we next tested the effect of the AT_{2} receptor blocker PD123319 (10^{-6} mol/L). With Ang II alone, increasing NaCl dilated preconstricted Af-Arts (maximal response: 10.6 ± 1.2 μm; EC_{50}: 16.6 ± 0.8 mmol/L). Adding PD123319 to the CNT perfusate had no effect on CTGF in the presence of Ang II (maximal response: 10.1 ± 1.0 μm; EC_{50}: 16.8 ± 1.0 mmol/L; Figure 2D). These data show that AT_{2} receptors do not mediate the effect of Ang II on CTGF.

Because PKC mediates at least some of the effects of Ang II along the nephron, we next tested the ability of the PKC inhibitor staurosporine to block the stimulatory effect of Ang II on CTGF. With Ang II alone, increasing NaCl dilated preconstricted Af-Arts (maximal response: 9.7 ± 1.5 μm; EC_{50}: 17.1 ± 1.3 mmol/L). Adding staurosporine (10^{-8} mol/L) to the CNT perfusate attenuated NaCl-induced dilation and the curve shifted to the right (maximal response: 7.2 ± 1.1 μm; EC_{50}: 30.9 ± 1.4 mmol/L; P<0.001 versus Ang II alone; Figure 3A). These data show that PKC mediates, at least in part, Ang II–induced potentiation of CTGF. In control experiments, we tested the effect of staurosporine on CTGF in the absence of exogenous Ang II. Staurosporine alone had no effect on CTGF. During the control period, the maximal response was 8.6 ± 1.0 μm and the EC_{50} was 36.9 ± 2.5 mmol/L. During staurosporine administration, the maximal response was 8.6 ± 1.2 μm and the EC_{50} was 35.4 ± 2.3 mmol/L (Figure 3B). These data suggest that the stimulatory effect of Ang II on CTGF is mediated by PKC activation.

Because we have reported that CTGF is initiated by activation of ENaC, we tested whether stimulation of CTGF by Ang II also requires ENaC activity. For this we added the selective ENaC blocker benzamil (10^{-6} mol/L) to the CNT perfusate along with Ang II. With Ang II alone, increasing NaCl dilated preconstricted Af-Arts. Adding benzamil completely blocked CTGF even in the presence of Ang II (Figure 4), suggesting that both CTGF and its stimulation by Ang II require ENaC activity.

**Discussion**

We investigated the regulation of CTGF by intratubular Ang II and found that adding Ang II to the CNT lumen enhances CTGF. These effects are mediated by AT_{1} but not AT_{2} receptors and require activation of PKC, because blocking PKC with staurosporine prevented Ang II from stimulating CTGF. We also established that ENaC activity is required for both CTGF and its stimulation by Ang II.

Ang II plays a key role in renal control of Na\(^+\), acid-base, and water balance, not only by controlling aldosterone secretion and affecting renal vasculature but also by acting directly on the nephron. Studies have shown that Ang II can alter ion transport in the distal tubular segment, cortical collecting duct, and macula densa. There is evidence that intratubular Ang II levels are much higher than those in the circulation and that intratubular Ang II is locally synthe-
sized, instead of filtered, suggesting some of the effects of Ang II on the nephron may be mediated by intratubular receptors. Furthermore, Ang II perfused in the lumen of the distal nephron can affect sodium and bicarbonate reabsorption. To our knowledge, the effect of intratubular Ang II in the CNT has not been specifically assessed, partly because perfusion of the CNT is difficult. However, data obtained in the collecting duct can provide some insight, because these 2 segments are similar. Wang and Giebisch showed that intratubular perfusion of Ang II stimulates Na transport in the collecting duct. Thus, we felt it was important to investigate whether intratubular Ang II directly regulates CTGF and the mechanism(s) involved. We found that adding Ang II to the CNT lumen enhanced CTGF and that, in the presence of a low, constant concentration of NaCl that does not induce CTGF per se, increasing intratubular Ang II concentrations in the lumen of the CNT dilated the Af-Art. This is the first report showing that Ang II can affect Af-Art hemodynamics by acting on the CNT.

Of note, the concentration of Ang II that we used in perfusing the CNT is well within the range of observed intratubular concentrations in the proximal tubule of normotensive animals. The concentration of Ang II in the CNT remains unknown because of technical difficulties. Estimations based on urinary concentrations range from 10^{-9} to 10^{-12} m. In addition, renin is known to be expressed in the CNT; therefore, it is possible that Ang II is generated locally. Ang II concentrations of 10^{-9} m were found to have an effect in the distal nephron, increasing ENaC activity. In our study, perfusion of the CNT with Ang II 10^{-10} m tended to potentiate CTGF, and further potentiation was seen with Ang II 10^{-9} m. Thus, the concentration that we used is similar to that reported to activate ENaC and within the range of previous estimates of distal Ang II concentration.

The actions of Ang II are mediated by 2 receptors, AT1 and AT2. Losartan can bind to and inactivate AT1, whereas PD123319 is commonly used as an antagonist for AT2. Most of the known physiological effects of Ang II in the adult kidney seem to be mediated by AT1, whereas the physiological role of AT2 has not been entirely clarified. Immuno- histochemical studies have established the existence of AT1 receptors at both the apical and basolateral membranes of distal nephron segments. We found that the stimulatory effects of intratubular Ang II on CTGF were prevented by the AT1 antagonist losartan but not the AT2 antagonist PD123319, suggesting that stimulation of CTGF by Ang II is mediated by the AT1 receptor.

The sequence of events that leads from occupying the membrane receptor to activation of transporters is not clear. Several studies have suggested that Ang II receptors in the renal epithelium are simultaneously coupled to a number of signal transduction pathways, including adenylyl cyclase, phospholipase A2, and phospholipase C/PKC. Du et al reported that Ang II regulates Na^+ absorption by a cAMP-independent mechanism and that PKC and intracellular calcium each play a critical role in modulating the effects of physiological concentrations of Ang II on proximal tubule transport. In medullary thick ascending limb suspensions, Ang II stimulates Na^+/K^+/2Cl^- cotransporter activity, and this effect was reportedly blocked by the PKC inhibitor staurosporine. PKC has also been shown to stimulate Na^+ uptake in the brush border membrane of proximal tubules. We, therefore, questioned whether PKC could be responsible for the stimulatory effect of Ang II on CTGF. We found that PKC mediates Ang II–induced enhancement of CTGF, because blocking PKC with staurosporine inhibited the stimulatory action of Ang II. Although the isoforms of PKC expressed in the CNT are unknown, there is evidence that both phorbol ester–sensitive and phorbol ester–insensitive isoforms are present in primary cultures of rabbit CNT and cortical collecting duct cells. Our findings clearly show the participation of PKC activity, which can be inhibited by staurosporine.

Here we report that activation of the AT1 receptors in the lumen of the CNT potentiates the vasodilatory effect of CTGF. This was not unexpected, because Peti-Peterdi et al found that Ang II stimulates ENaC activity, and this channel mediates CTGF. We found that benzamid competitively blocked CTGF in the presence of Ang II. Previously we have reported that blockade of ENaC completely blocks CTGF; thus, these data suggest that the presence of ENaC activity is necessary for both CTGF and its potentiation by Ang II.

The present study further characterizes the newly described CTGF mechanism. CTGF emerges as a potential regulator of Af-Art tone that may serve to counter circulating vasoconstrictors. In this study we show that CTGF is not only able to dilate NE-preconstricted arterioles but also Ang II–preconstricted arterioles, indicating that CTGF is not specific for NE but may counter other vasoconstrictors. In addition, because we also show that intratubular Ang II potentiates vasodilatory CTGF, these data indicate that Ang II in different renal compartments regulates Af-Art tone differently.

Ang II, like many, if not all, prohypertensive mediators, also has some mechanisms that limit its hypertensive effect. Such mechanisms include acting in receptors with different actions (eg, activation of vasoconstrictor AT1 receptors and vasorelaxant AT2 receptors), simultaneously stimulating mediators with opposing effects (eg, the stimulation by Ang II of both O2^- and NO production in the thick ascending limb), generation of a metabolite with opposing effects (eg, Ang I-7 derived from the action of angiotensin-converting enzyme 2), or having different effects in different compartments. An example of the latter is stimulation of renin release in the distal nephron by intratubular Ang II, which opposes the actions of Ang II in the vasculature (inhibition of renin release from the Af-Art). Thus, it is not surprising that Ang II, via CTGF, may potentiate a mechanism that limits vasoconstriction of the Af-Art.

In summary, using an isolated perfused Af-Art with the CNT attached we found that intratubular Ang II augmented CTGF response. Adding losartan to the CNT lumen completely blocked the effect of Ang II, whereas losartan by itself had no effect on CTGF. PD123319 in the CNT lumen did not influence the effect of Ang II on CTGF. Activation of PKC is responsible for the stimulatory effect of Ang II. Both CTGF and its stimulation by Ang II require ENaC.
Perspectives

CTGF is a novel regulatory mechanism of the renal microcirculation. It could account in part for the Af-Art dilation and increased GFR observed during high-salt intake or volume expansion, by antagonizing or resetting TGF and favoring Na excretion. Intratubular Ang II, acting on AT1 receptors in the CNT, potentiates CTGF. This may be particularly important in the setting of volume expansion, where intratubular Ang II levels are much higher than circulating ones.15

For a long time, the function of Ang II has been known to cause vasoconstriction with relative preservation of GFR as compared with other vasoconstrictors.48,49 This has been attributed to the differential sensitivity of afferent and efferent arterioles to the action of Ang II with the afferent constricting relatively less than the efferent. Our present study is the first to suggest that potentiation of CTGF by intratubular Ang II could serve to limit Af-Ar vasoconstriction induced by circulating Ang II, thus preserving GFR and preventing glomerular damage.

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Disclosures
None.

References
14. Miyata N, Kauffman S, Katz AI. Angiotensin II binding sites in indi-

40. Zhao D, Seth DM, Navar LG. Enhanced distal nephron sodium reab-

angiotensin receptors: a report of the Nomenclature Committee of the

42. Bouby N, Hus-Citharel A, Marchetti J, Bankir L, Corvol P, Llorens-
Cortes C. Expression of type 1 angiotensin II receptor subtypes and

43. Harrison-Bernard LM, Navar LG, Ho MM, Vinson GP, El-Dahr SS.
Immunohistochemical localization of ANG II AT1 receptor in adult rat

44. Wang T, Chan YL. Mechanism of angiotensin II action on proximal

45. Liu F-Y, Cogan MG. Role of protein kinase C in proximal bicarbonate

46. Du Z, Ferguson W, Wang T. Role of PKC and calcium in modulation of
effects of angiotensin II on sodium transport in proximal tubule. *Am J

47. van BJ, Hoenderop JG, Groenendijk M, Van Os CH, Bindels RJ, Willems
PH. Hormone-stimulated Ca2+ transport in rabbit kidney: multiple sites

48. Ichikawa I, Harris RC. Angiotensin actions in the kidney: renewed insight

49. Ito S, Arima S, Juncos LA, Carretero OA. Endothelium-derived relaxing
factor/nitric oxide modulates angiotensin II action in the isolated
microperfused rabbit afferent but not efferent arteriole. *J Clin Invest.*
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