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Renal denervation has blood pressure–independent protective effects on kidney and heart in a rat model of chronic kidney disease

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We elucidate the underlying mechanisms of bidirectional cardiorenal interaction, focusing on the sympathetic nerve driving disruption of the local renin–angiotensin system (RAS). A rat model of N^{ω}-nitro-L-arginine methyl ester (L-NAME; a nitric oxide synthase inhibitor) administration was used to induce damage in the heart and kidney, similar to cardiorenal syndrome. L-NAME induced sympathetic nerve–RAS overactivity and cardiorenal injury accompanied by local RAS elevations. These were suppressed by bilateral renal denervation, but not by hydralazine treatment, despite the blood pressure being kept the same between the two groups. Although L-NAME induced angiotensinogen (AGT) protein augmentation in both organs, AGT mRNA decreased in the kidney and increased in the heart in a contradictory manner. Immunostaining for AGT suggested that renal denervation suppressed AGT onsite generation from activated resident macrophages of the heart and circulating AGT excretion from glomeruli of the kidney. We also examined rats treated with L-NAME plus unilateral denervation to confirm direct sympathetic regulation of intrarenal RAS. The levels of urinary AGT and renal angiotensin II content and the degrees of renal injury from denervated kidneys were less than those from contralateral innervated kidneys within the same rats. Thus, renal denervation has blood pressure–independent beneficial effects associated with local RAS inhibition.

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Chronic kidney disease (CKD) may lead to heart failure and is associated with high mortality,[1] and heart failure may lead to renal failure.[2] Cardiac and renal dysfunction amplifies the failure of the other organ in acute and/or chronic situations. A new classification[3] was proposed to help the 'clinical' understanding of heart and kidney interactions, but understanding of the 'biological' mechanisms of this interaction remains limited. The sympathetic nervous system, the renin–angiotensin system (RAS), and nitric oxide (NO) are thought to have key roles in the cardiorenal cycle.[4] To investigate sympathetic nerve–RAS interaction, we developed a cardiorenal syndrome model with induced chronic NO depletion.

Although several clinical studies have reported the potential efficacies of renal sympathetic denervation (DNx) for resistant hypertension,[5] left ventricular hypertrophy,[6] albuminuria,[7] glucose metabolism,[8] sleep apnea syndrome,[9] arterial stiffness,[10] and CKD,[11] most studies were uncontrolled. A recent study, the SYMPLICITY-HTN-3 trial, was a blinded, randomized sham-controlled trial and failed to show any benefit of catheter-based DNx.[12] Currently, the underlying mechanisms are unclear and information on the procedure is limited.[13] Many clinicians think the clinically refractory nature of cardiorenal syndrome is an attractive target for intervention. The current study examined the beneficial effects of DNx on the sympathetic cross-talk with RAS in cardiorenal syndrome, with a focus on local RAS in the heart and kidney.

Local RAS has paracrine and intracrine roles independent of circulating RAS, and have been extensively reviewed.[14–17] Physiologically, intrarenal RAS regulates the glomerular filtration rate,[18] blood pressure, and proximal tubular reabsorption.[19] Brain RAS modulates drinking behavior[20] and blood pressure.[21] Cardiac RAS has inotropic and chronotropic effects.[22] Pathophysiologically, a long-term elevation in local RAS leads to organ dysfunction: for example, hypertension and progression of CKD in the kidney,[23–25] hypertension in the brain,[26] cardiac hypertrophy, fibrosis, and remodeling in the heart.[27] Despite vast amounts of research on the local regulation of RAS, its mechanisms remain controversial.[28–31] This study also addressed the

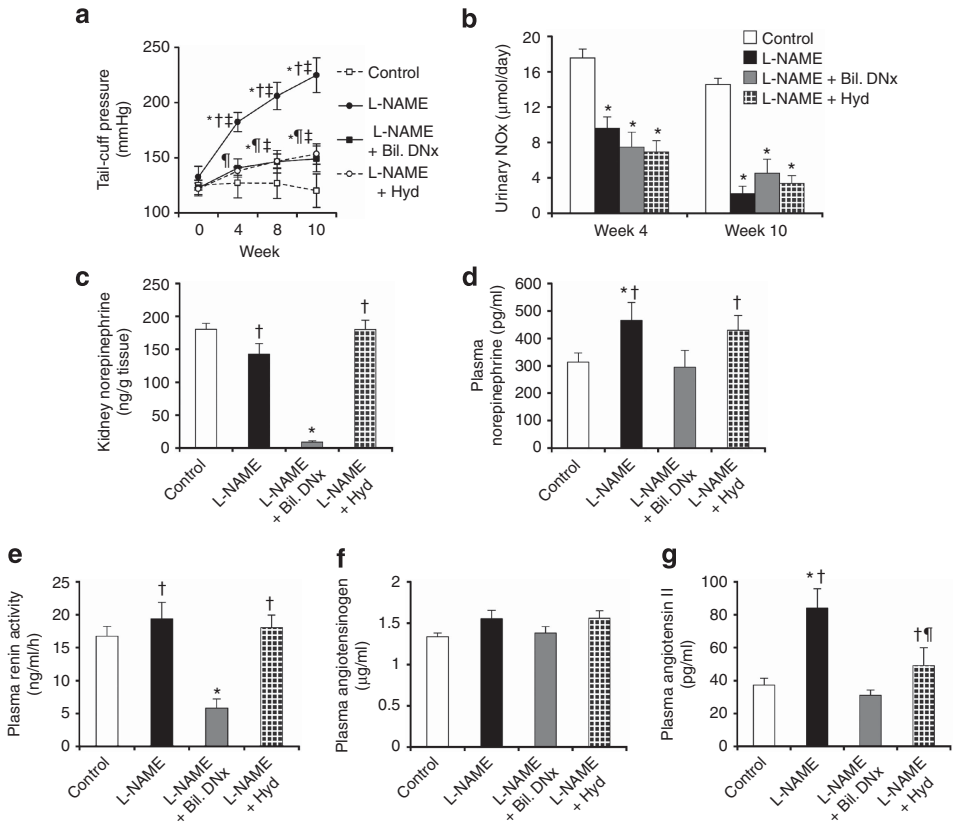

Figure 1 | The validity of renal denervation, blood pressure, and circulating factors in protocol 1. (**a**) Serial measurements of tail-cuff pressure. (**b**) Urinary nitrate/nitrite. Nitric oxide metabolite (NOx) levels at weeks 4 and 10 were equally suppressed among the three groups. (**c**) Renal cortical norepinephrine content levels were measured to confirm the validity of renal denervation. (**d**) Plasma norepinephrine and (**e**–**g**) RAS components were measured at the end of the study (week 10). Values are mean ± s.e.m. *$P<0.05$ vs. control rats, $^{\dagger}P<0.05$ vs. Bil. DNx rats, $^{\P}P<0.05$ vs. L-NAME rats, $^{\ddagger}P<0.05$ vs. baseline values (week 0). Bil. DNx, bilateral renal denervation; L-NAME; N^{ω}-nitro-L-arginine methyl ester.

currently highly debated issue of the origin (tubular origin[31] or leakage from glomeruli[30]) of intrarenal angiotensinogen (AGT).

RESULTS

Renal denervation drives blood pressure–independent cardiorenal protection

We employed a cardiorenal syndrome model with NO depletion, which induced CKD and heart failure and bidirectional interaction between both damaged organs. In protocol 1, chronic N^{ω}-nitro-L-arginine methyl ester (L-NAME) administration induced a progressive elevation in tail-cuff pressure that reached approximately 220 mmHg over 10 weeks, and also showed lower body weights and hypoalbuminemia at the end of the study compared with other groups (Supplementary Table S1 online). Bilateral renal denervation (Bil. DNx) ameliorated the severe hypertension induced by L-NAME, resulting in tail-cuff pressure of approximately 150 mmHg at week 10. Tail-cuff pressures in rats treated with L-NAME and hydralazine were comparable to those in Bil. DNx rats (Figure 1a). These differences among L-NAME-treated groups were observed despite equal suppression of urinary and tissue NO metabolite production (Figure 1b and Supplementary Table S1 online). DNx was confirmed by a marked decrease in norepinephrine content in the denervated kidney at week 10 (Figure 1c). Systemic sympathetic nerve activity was inferred from plasma and urinary norepinephrine levels at week 10 (Figure 1d and Supplementary Table S1 online). These elevations were suppressed by Bil. DNx, but not by hydralazine (Figure 1d). Plasma renin activity was markedly suppressed in Bil. DNx rats compared with the other groups (Figure 1e). Plasma angiotensin II (AII) levels were higher in L-NAME- or hydralazine-treated rats compared with control or Bil. DNx rats (Figure 1g). There were no significant differences in plasma AGT levels, liver AGT protein, or mRNA levels among the groups (Figure 1f and Supplementary Figure S1 online). L-NAME administration induced severe proteinuria, glomerulosclerosis, and interstitial fibrosis (Figure 2a–e) and elevation in serum creatinine and blood urea nitrogen levels (Supplementary Table S1 online). Treatment with Bil. DNx, but not with hydralazine, suppressed all these changes in the kidney (Figure 2a–e), despite blood pressure being the same between these two groups (Figure 1a). L-NAME administration also induced cardiac hypertrophy, fibrosis, and systolic dysfunction. L-NAME-treated rats showed increased heart weight corrected by body weight (Figure 3a) and had left ventricular hypertrophy accompanied by cardiac fibrosis