Discoidin domain receptor 1 kinase activity is required for regulating collagen IV synthesis

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Abstract

Discoidin domain receptor 1 (DDR1) is a receptor tyrosine kinase that binds to and is activated by collagens. DDR1 expression increases following kidney injury and accumulating evidence suggests that it contributes to the progression of injury. To this end, deletion of DDR1 is beneficial in ameliorating kidney injury induced by angiotensin infusion, unilateral ureteral obstruction, or nephrotoxic nephritis. Most of the beneficial effects observed in the DDR1-null mice are attributed to reduced inflammatory cell infiltration to the site of injury, suggesting that DDR1 plays a pro-inflammatory effect. The goal of this study was to determine whether, in addition to its pro-inflammatory effect, DDR1 plays a deleterious effect in kidney injury by directly regulating extracellular matrix production. We show that DDR1-null mice have reduced deposition of glomerular collagens I and IV as well as decreased proteinuria following the partial renal ablation model of kidney injury. Using mesangial cells isolated from DDR1-null mice, we show that these cells produce significantly less collagen compared to DDR1-null cells reconstituted with wild type DDR1. Moreover, mutagenesis analysis revealed that mutations in the collagen binding site or in the kinase domain significantly reduce DDR1-mediated collagen production. Finally, we provide evidence that blocking DDR1 kinase activity with an ATP-competitive small molecule inhibitor reduces collagen production. In conclusion, our studies indicate that the kinase activity of DDR1 plays a key role in DDR1-induced collagen synthesis and suggest that blocking collagen-mediated DDR1 activation may be beneficial in fibrotic diseases.

Introduction

Discoidin Domain Receptor 1 (DDR1) is a receptor tyrosine kinase that binds collagens [1] and has been implicated in the regulation of multiple cellular functions including migration [2–5], cytokine secretion [6–8], and extracellular matrix homeostasis/remodeling [9–11]. DDR1 is required for normal tissue development, but the function of DDR1 in adult tissues particularly in diseased tissues is poorly understood [12,13]. DDR1 contributes to cancer [14,15] and promotes inflammation in models of atherosclerosis [10,16] and lung fibrosis [17], but the mechanisms whereby DDR1 contributes to disease progression are not clear.

DDR1 upregulation in patients with kidney diseases such as lupus nephritis and Goodpasture's syndrome [8] as well as in animal models of kidney injury [7,8,18] suggests that DDR1 plays an important role in kidney disease. Studies in various mouse models of kidney injury indicate that DDR1 deletion results in improved renal function and reduced

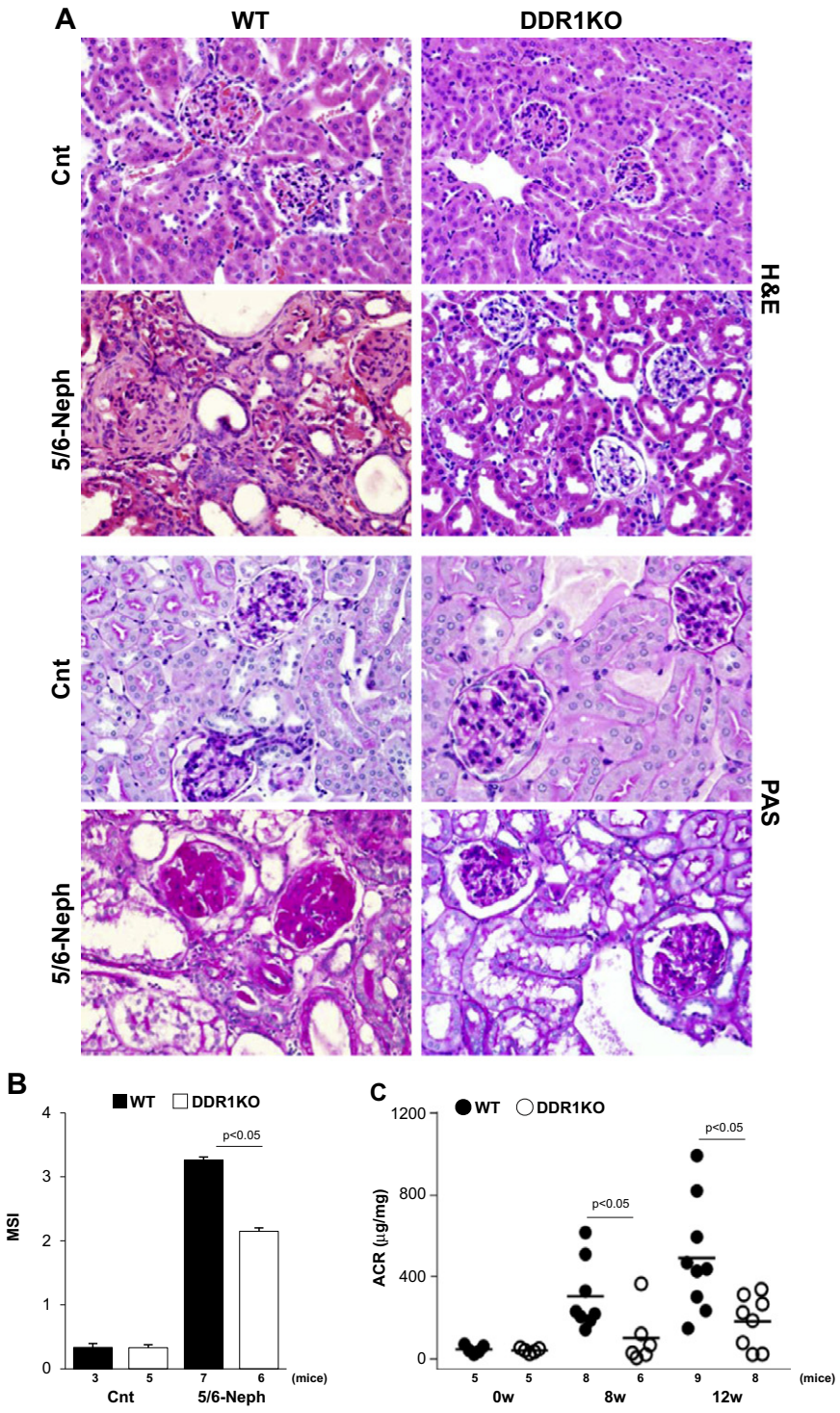

Fig. 1. Loss of DDR1 reduces partial renal ablation-mediated glomerular injury. Kidney injury was induced by performing partial renal ablation (5/6 Nephr) in wild type (WT) and DDR1KO mice. Mice were sacrificed 12 weeks after injury and kidneys were collected for histological analysis. (A) Representative light micrographs of Hematoxylin and Eosin (H&E) and periodic-acid-Schiff (PAS) stained kidneys from uninjured (Cnt) or 12 weeks post 5/6 Nephr. Loss of DDR1 rescues the severe glomerular damage observed in injured WT mice. Original magnification, 40×. (B) Mesangial sclerosis index (MSI) was evaluated 12 weeks after 5/6 Nephr and scored as described in the Methods. Values represent the mean ± SEM of the number of mice indicated. (C) Urine albumin excretion, expressed as albumin-to-creatinine ratio (ACR), was examined on the number of mice indicated at 0, 8, and 12 weeks after 5/6 Nephr. Mice lacking DDR1 show significantly less proteinuria following injury.