[Frontiers in Bioscience E2, 1246-1249, June 1, 2010]

Role of (pro)renin receptor in cardiovascular cells from the aspect of signaling

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1. ABSTRACT

The renin-angiotensin-aldosterone system regulates homeostasis of salt and water, vasoconstriction, and remodeling in cardiovascular and renal cells via activation of intracellular signaling pathway. Prorenin, the precursor of renin, had long been considered to be an inactive form. However, a receptor—the (pro)renin receptor—that binds to both renin and prorenin has been recently identified. Prorenin binding to (pro)renin receptors both results in angiotensinogen cleaving into angiotensin (Ang) I, and triggers activation of (pro)renin receptor-stimulated signal transduction pathways, independent of generating Ang II. In the last decade, it has been reported that the intracellular signaling pathway is activated by prorenin in cardiomyocytes, mesangial cells, podocytes, distal tubular cells, vascular endothelial cells and vascular smooth muscle cells, indicating that prorenin mediates intracellular effects in various cardiovascular and kidney cells. In this review, we summarize novel intracellular signaling systems and their downstream effects via (pro)renin receptors in cardiovascular and kidney cells.

2. INTRODUCTION

Prorenin, the precursor of renin, has been considered to be an inactive form, despite its high concentration in plasma relative to renin levels (1). Recently, a receptor for both prorenin and renin—hereinafter referred to as the (pro)renin receptor or (P)RR—was identified (2). The binding of prorenin to the (P)RR results in angiotensinogen cleaving into angiotensin (Ang) peptides, and does so with kinetics similar to those of active renin with attached prosegments (2). Activation of (P)RR by renin or prorenin binding also triggers activation of (P)RR-stimulated signal transduction pathways, independent of generating Ang II (2-4). In the last decade, it has been reported that (P)RR activation, independent of Ang II action, can induce activation of intracellular signaling pathway in cardiomyocytes (3), mesangial cells (2), podocytes (5), distal tubular cells (6), vascular endothelial cells (7), vascular smooth muscle cells (8), and monocytes (9), indicating that prorenin mediates intracellular effects in various cardiovascular and kidney cells. In this short review, we focus on novel intracellular

signaling systems via (P)RRs in cardiovascular and kidney cells.

3. TISSUE TYPES

3.1. Heart

Transgenic rats that overexpress rat prorenin develop cardiac injuries, as evidenced by hypertrophic cardiomyocytes, and subendocardial and pericoronary fibrosis without hypertension (10). This indicates that prorenin may affect hypertrophic cardiac alterations much as it reportedly affects cardiac hypertrophy and heart cell proliferation via activation of intracellular signaling pathway (3, 11). Northern blot analysis has detected (P)RR mRNA in adult human heart tissue (2); protein expression of (P)RR has been observed in coronary artery smooth muscle tissue (2) and cultured neonatal cardiomyocytes (3). Prorenin is reportedly also involved in intracellular signaling in cardiomyocytes (3, 11). Prorenin simultaneously induces phosphorylation of both p38 mitogen-activated protein (MAP) kinase and HSP27 (the latter is involved in maintaining cell growth, survival and structural integrity) in cardiomyocytes, although prorenin affects neither phosphorylation of extracellular regulated kinase (ERK) 1/2 nor release of plasminogen-activator inhibitor-1 (PAI-1) (3). As Ang type1 (AT_1) receptor antagonists do not block phosphorylation of MAP kinases and subsequent transcriptional effects by prorenin, these effects are considered independent of Ang II. (Pro)renin may induce cardiac hypertrophy in rats that overexpress prorenin, independent of Ang II generation via (P)RR-mediated MAP kinase activation.

3.2. Kidneys

Prorenin also induces renal injury in transgenic rats that overexpress prorenin (10). Pathological kidney lesions in these rats are consistent with moderate to severe nephropathy and angiopathy such as glomerulosclerosis, tubulointerstitial atrophy and inflammation, and thickened arterial walls. (P)RRs are expressed in kidneys; however, their expression is lower than in brain, heart or pancreas (2). Immunofluorescent staining of kidney cortex using anti-(P)RR antibody showed receptor expression in glomeruli and vasculature (2), and confirmed that labeled (P)RRs were found in the mesangium of glomeruli. Receptor-mediated actions induced by renin or prorenin are also reported in cultured mesangial cells (MCs) (2, 12). The binding of renin to functional cell-surface receptors increased ^{3}H-thymidine incorporation (an index of cell proliferation), and PAI-1 antigen production, independently of renin enzyme activity (12). Renin binding also induced ERK 1/2 activation in transfected human MCs that overexpressed human (P)RRs (2). Importantly, renin binding does not alter intracellular calcium or cyclic AMP, suggesting that MAP kinase activation is a critical signaling pathway for (P)RR in MCs. The presence of angiotensin-converting enzyme (ACE) inhibitors and AT_1 receptor antagonists confirms that renin-induced MAP kinase activation is independent of any Ang II generation or action.

Later studies demonstrated the upstream and downstream effects of MAP kinase phosphorylation (13-16). Renin induces cell proliferation along with transforming growth factor (TGF)-β via MAP kinase phosphorylation in MCs (13). Renin increases TGF-β expression and matrix proteins such as PAI-1, fibronectin and type 1 collagen in MCs (14). These effects are not altered by adding an inhibitor of renin's enzymatic action (RO 42-5892), a AT_1 receptor antagonist or an ACE inhibitor. However, (P)RR siRNA inhibited these effects, indicating that renin induces TGF-β production through a receptor-mediated mechanism, independent of Ang II generation or action. Prorenin can also induce genomic actions which are involved in both TGF-β-dependent and TGF-β-independent pathways, and contributes to proinflammatory and profibrotic protein expression in MCs (15). Both (P)RR siRNA and handle region peptide (HRP) —which acts as a prorenin decoy—attenuated ERK 1/2 phosphorylation, type IV collagen release and TGF-β mRNA in MCs (16). Furthermore, MCs synthesize and secrete prorenin and renin (16). Recently, glucose-induced (P)RR up-regulation was reported; D-glucose increases prorenin, renin and (P)RR expressions, and augments phosphorylation of ERK 1/2, c-Jun N-terminal kinase (JNK), c-Jun and nuclear factor-κB (17).

Although (P)RR expression was initially reported in mesangium (2), subsequent reports indicate that it is also expressed in podocytes (5, 18), distal tubules and collecting ducts (6). Expression of (P)RRs in podocytes at the protein level were confirmed in vivo, and was also confirmed in cultured podocytes at mRNA levels. In addition, glomerular upregulation of (P)RR in podocytes was observed in kidneys of rats with early diabetic nephropathy (5), indicating that podocyte injury seen in diabetic nephropathy might be involved in prorenin-(P)RR binding. On the other hand, some (P)RR functions were elucidated using Madin-Darby canine kidney (MDCK) cells, which are collecting duct/distal tubule cells (6). (Pro)renin induces ERK 1/2 phosphorylation in MDCK cells in presence of both AT_1 receptor antagonist and Ang II type 2 receptor antagonist; phosphorylation is inhibited by (P)RR siRNA, suggesting that these effects are Ang II-independent, and receptor-mediated. Induction of MAP kinase activation by (pro)renin has been shown to be related to vacuolar H^+-ATPase in MDCK cells. In human embryo kidney (HEK) 293 cells, downstream actions of (P)RR activation reportedly include decreased (P)RR mRNA, increased phosphatidylinositol-3 (PI3) kinase-p85α mRNA, and augmented viable cell numbers (19). Aliskiren, a direct inhibitor of renin, does not affect noncatalytic effects of (pro)renin on HEK293 cells. These findings suggest a possible mechanism of (pro)renin-induced renal injury, independent of Ang II generation, as reported in *in vivo* experiments (10).

3.3. Vasculature

(P)RR mRNA and protein expression are seen in vasculature, including vascular smooth muscle cells (VSMCs) (8) and endothelial cells (ECs) (7). Aortic wall hypertrophy is seen in prorenin-transgenic rats without high blood pressure (10). In addition to animal model experiments, intracellular signaling activation induced by

(pro)renin is also reported in vascular cells. Activation of MAP kinase was shown in human VSMCs (8), similarly to cardiac and kidney cells. Prorenin induces phosphorylation of ERK 1/2 through Ang II-independent, (P)RR-mediated activation of tyrosine kinase and subsequent ERK kinase in human VSMCs. However, neither p38 MAP kinase nor JNK is activated by incubation with prorenin in VSMCs. Furthermore, not only did AT_1 receptor antagonist and ACE inhibitor fail to attenuate (pro)renin-induced ERK phosphorylation, but so did HRP and the direct renin inhibitor, aliskiren (20). Although catalytic activities of prorenin and renin could be blocked by renin inhibitor, direct (P)RR signaling might not be inhibited by aliskiren or HRP in VSMCs. Subsequent MAP kinase phosphorylation showed that (pro)renin increased ^{3}H-thymidine incorporation into VSMC (4), and PAI-1 mRNA expression (21). PAI-1 protein expression is also upregulated by prorenin, which is attenuated by (P)RR siRNA, but not by AT_1 receptor antagonist, indicating that upregulation is a receptor-mediated effect of prorenin. Recently, we showed that prorenin-induced MAP kinase activation required epidermal growth factor receptor phosphorylation. Furthermore, PI3 kinase-Akt pathway is activated by prorenin, and subsequently induces cell hypertrophy and proliferation in rat VSMCs—suggesting a molecular mechanism of prorenin-induced hypertrophic alteration in vasculature. (P)RRs were also expressed not only in VSMCs but also in vascular ECs (7). RT-PCR analysis revealed that ECs from various vascular beds such as umbilical vein, lung and coronary artery express (P)RR mRNA. Immunoblotting analysis also confirmed the expression of (P)RRs at protein level. Prorenin induces ERK 1/2 phosphorylation, and plays an important role in regulation of endothelial function, including proliferation, migration, tube formation and apoptosis.

4. CONCLUSION

Prorenin binding to (P)RR promotes two distinct mechanisms: (a) Ang II generation induced by nonproteolytic activation of prorenin; and (b) activation of (P)RR-mediated Ang II-independent signaling pathways. In this review, we summarized the findings that (P)RR activation, independent of Ang II, can induce MAP kinase phosphorylation in cardiovascular and kidney cells. Intracellular signaling activated by prorenin and renin may be involved in organ injury. The (P)RR-dependent, but Ang II-independent, effects of prorenin provide new insights into mechanisms of cardiovascular and renal cells, and novel therapeutic targets.

5. REFERENCES

1. F. H. Derkx and M. A. Schalekamp: Human prorenin: pathophysiology and clinical implications. *Clin Exp Hypertens A*, 10(6), 1213-25 (1988)

2. G. Nguyen, F. Delarue, C. Burckle, L. Bouzhir, T. Giller and J. D. Sraer: Pivotal role of the renin/prorenin receptor in angiotensin II production and cellular responses to renin. *J Clin Invest*, 109(11), 1417-27 (2002)

3. J. J. Saris, P. A. t Hoen, I. M. Garrelds, D. H. Dekkers, J. T. den Dunnen, J. M. Lamers and A. H. Jan Danser: Prorenin induces intracellular signaling in cardiomyocytes independently of angiotensin II. *Hypertension*, 48(4), 564-71 (2006)

4. W. W. Batenburg, R. J. de Bruin, J. M. van Gool, D. N. Muller, M. Bader, G. Nguyen and A. H. Danser: Aliskiren-binding increases the half life of renin and prorenin in rat aortic vascular smooth muscle cells. *Arterioscler Thromb Vasc Biol*, 28(6), 1151-7 (2008)

5. R. V. Durvasula and S. J. Shankland: Activation of a local renin angiotensin system in podocytes by glucose. *Am J Physiol Renal Physiol*, 294(4), F830-9 (2008)

6. A. Advani, D. J. Kelly, A. J. Cox, K. E. White, S. L. Advani, K. Thai, K. A. Connelly, D. Yuen, J. Trogadis, A. M. Herzenberg, M. A. Kuliszewski, H. Leong-Poi and R. E. Gilbert: The (Pro)renin receptor: site-specific and functional linkage to the vacuolar H+-ATPase in the kidney. *Hypertension*, 54(2), 261-9 (2009)

7. M. Uraoka, K. Ikeda, Y. Nakagawa, M. Koide, Y. Akakabe, R. Nakano-Kurimoto, T. Takahashi, S. Matoba, H. Yamada, M. Okigaki and H. Matsubara: Prorenin induces ERK activation in endothelial cells to enhance neovascularization independently of the renin-angiotensin system. *Biochem Biophys Res Commun*, 390(4), 1202-7 (2009)

8. M. Sakoda, A. Ichihara, Y. Kaneshiro, T. Takemitsu, Y. Nakazato, A. H. Nabi, T. Nakagawa, F. Suzuki, T. Inagami and H. Itoh: (Pro)renin receptor-mediated activation of mitogen-activated protein kinases in human vascular smooth muscle cells. *Hypertens Res*, 30(11), 1139-46 (2007)

9. S. Feldt, W. W. Batenburg, I. Mazak, U. Maschke, M. Wellner, H. Kvakan, R. Dechend, A. Fiebeler, C. Burckle, A. Contrepas, A. H. Jan Danser, M. Bader, G. Nguyen, F. C. Luft and D. N. Muller: Prorenin and renin-induced extracellular signal-regulated kinase 1/2 activation in monocytes is not blocked by aliskiren or the handle-region peptide. *Hypertension*, 51(3), 682-8 (2008)

10. M. Veniant, J. Menard, P. Bruneval, S. Morley, M. F. Gonzales and J. Mullins: Vascular damage without hypertension in transgenic rats expressing prorenin exclusively in the liver. *J Clin Invest*, 98(9), 1966-70 (1996)

11. J. J. Saris, M. M. van den Eijnden, J. M. Lamers, P. R. Saxena, M. A. Schalekamp and A. H. Danser: Prorenin-induced myocyte proliferation: no role for intracellular angiotensin II. *Hypertension*, 39(2 Pt 2), 573-7 (2002)

12. G. Nguyen, F. Delarue, J. Berrou, E. Rondeau and J. D. Sraer: Specific receptor binding of renin on human mesangial cells in culture increases plasminogen activator inhibitor-1 antigen. *Kidney Int*, 50(6), 1897-903 (1996)

13. Y. Huang, N. A. Noble, J. Zhang, C. Xu and W. A.

Border: Renin-stimulated TGF-beta1 expression is regulated by a mitogen-activated protein kinase in mesangial cells. *Kidney Int*, 72(1), 45-52 (2007)

14. Y. Huang, S. Wongamorntham, J. Kasting, D. McQuillan, R. T. Owens, L. Yu, N. A. Noble and W. Border: Renin increases mesangial cell transforming growth factor-beta1 and matrix proteins through receptor-mediated, angiotensin II-independent mechanisms. *Kidney Int*, 69(1), 105-13 (2006)

15. R. A. Melnyk, J. Tam, Y. Boie, B. P. Kennedy and M. D. Percival: Renin and prorenin activate pathways implicated in organ damage in human mesangial cells independent of angiotensin II production. *Am J Nephrol*, 30(3), 232-43 (2009)

16. M. He, L. Zhang, Y. Shao, X. Wang, Y. Huang, T. Yao and L. Lu: Inhibition of renin/prorenin receptor attenuated mesangial cell proliferation and reduced associated fibrotic factor release. *Eur J Pharmacol*, 606(1-3), 155-61 (2009)

17. J. Huang and H. M. Siragy: Glucose promotes the production of interleukine-1beta and cyclooxygenase-2 in mesangial cells via enhanced (Pro)renin receptor expression. *Endocrinology*, 150(12), 5557-65 (2009)

18. A. Ichihara, Y. Kaneshiro, T. Takemitsu, M. Sakoda and H. Itoh: The (pro)renin receptor and the kidney. *Semin Nephrol*, 27(5), 524-8 (2007)

19. J. H. Schefe, C. Neumann, M. Goebel, J. Danser, S. Kirsch, R. Gust, U. Kintscher, T. Unger and H. Funke-Kaiser: Prorenin engages the (pro)renin receptor like renin and both ligand activities are unopposed by aliskiren. *J Hypertens*, 26(9), 1787-94 (2008)

20. S. Feldt, U. Maschke, R. Dechend, F. C. Luft and D. N. Muller: The putative (pro)renin receptor blocker HRP fails to prevent (pro)renin signaling. *J Am Soc Nephrol*, 19(4), 743-8 (2008)

21. J. Zhang, N. A. Noble, W. A. Border, R. T. Owens and Y. Huang: Receptor-dependent prorenin activation and induction of PAI-1 expression in vascular smooth muscle cells. *Am J Physiol Endocrinol Metab*, 295(4), E810-9 (2008)

Abbreviations: (P)RR: (pro)renin receptor; Ang: angiotensin; MAP: mitogen-activated protein; ERK: extracellular regulated kinase; PAI-1: plasminogen-activator inhibitor-1; AT_1: angiotensin type1; MC: mesangial cell; ACE: angiotensin-converting enzyme; TGF: transforming growth factor; HRP: handle region peptide; JNK: c-Jun N-terminal kinase; MDCK: Madin-Darby canine kidney; HEK: human embryo kidney; PI3: phosphatidylinositol-3; VSMC: vascular smooth muscle cell; EC: endothelial cell

Key Words: prorenin, (pro)renin receptor, intracellular signaling, MAP kinase, TGF-beta, Review

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