Analysis on the mechanism of reduced nephron number and the pathological progression of chronic renal failure in Astrin deficient rats

Abstract of Doctoral Thesis

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Abstract

Chronic kidney disease (CKD) is a common disease exhibiting globally high morbidity rate. Dialysis patient suffered from CKD is annually increasing because, except for dialysis and transplantation therapies, any effective treatment for CKD has not yet been developed. Moreover, considerable attention is focused on CKD as a risk factor for cardiovascular disease with highly lethality. Therefore, the elucidation of CKD pathogenesis, the identification of surrogate markers, and the development of effective treatment are required. Although CKD is caused by various congenital and acquired factors, there is a common pathological process in which renal lesions irreversibly progress due to reduced nephron number causing overload to remaining nephron (Brenner's theory). Progressive reduction of nephron in this process results in renal anemia and fibrosis at end-stage kidney disease. In such a background, it has been believed that low nephron number at birth is a risk for pathogenesis and prognosis of CKD. Revealing the mechanism by which nephron number is determined during renal development will provide crucial information useful for developing kidney regeneration therapy and for estimating a risk for CKD after birth. In addition, understanding the pathological mechanism by which CKD progresses under congenitally reduced nephron number will provide clues to inhibit the pathogenesis common among CKDs caused by various factors. In the present study, I analyzed the cause of reduced nephron number during embryonic development and the pathological progression of CKD in hypoplastic kidney (HPK) rats with congenitally reduced number of nephron resulting from loss-of-function type mutation of Astrin gene. Finally I discussed about possible therapeutic reagents based on results obtained. At first I demonstrated that HPK rats exhibit macrocytic erythropenia, normal plasma erythropoietin (EPO) levels accompanied by increased hepatic EPO production, and splenic hemosiderosis accompanied by a tendency toward

increased hemolysis during the progression of CKD. Then I revealed that HPK rats show glomeruloscrelosis triggered by podocyte injury and interstitial fibrosis accompanied by increased myofibroblasts during the progression of CKD with age, indicating congenital 80% nephron reduction causes progressive CKD resulting in renal fibrosis in rats. In studies of embryonic stage, I found that Astrin is expressed in metanephric mesenchyme (MM) cells derived from normal metanephors. Furthermore *in vivo* and *in vitro* (organ culture and cell culture) experiments using HPK metanephros revealed that the loss of Astrin causes thinning of nephrogenic layer due to altered stemness, increased apoptosis, and decreased proliferation in MM cells. These defects cause decreased branching of ureteric bud and early exhaustion of MM cells, finally leading to reduced number of nephron. Interestingly my experiments also indicated that these defective processes are involved in abnormal activation of mTOR signaling. Finally, my collaborating research demonstrates that mTOR inhibitor, Everolimus, can attenuate renal fibrosis and dysfunction in HPK rats. Taken together, through a series of my studies, it was indicated that activation of mTOR signaling is associated with not only fibrosis via increased myofibroblasts but also early loss of mesenchyme nephrogenic progenitors. I hope in future these evidences will contribute to reveal congenital risk for CKD, to improve renal regeneration therapies, and to develop molecularly targeted drag therapies against renal fibrosis.