Cell and molecular biology and mechanisms of early nephrogenesis.

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Editorial

The development of mammalian kidney involves several basic developmental processes, including branching morphogenesis, epithelial cell polarization and epithelial-mesenchymal transition. It is initiated when the intermediate mesoderm-derived Wolffian duct (WD) grows caudally, induces the formation of pronephric and mesonephric kidney and later forms the ureteric bud (UB) through inductive interaction from the metanephric mesenchyme (MM) and renal stroma [1]. Murine UB outgrows from the WD at E10.5, invades the MM and branches to form a tree-like tubular structure of the urinary collecting system starting from E11.5 [2].

Renal organogenesis is tightly regulated by several signaling pathways, and is launched when the UB outgrows from the Wolffian duct and invades the neighboring specified, but uninduced metanephric mesenchyme [1,3]. Glial cell line-derived neurotrophic factor (*Gdnf*) is expressed in the MM adjacent to the caudal WD and later in the MM surrounding the UB tips. Together with its c-Ret receptor, *Gdnf* plays a crucial role in kidney development [4]. Inactivation of *Gdnf*, Ret or its co-receptor *Gfrα1* leads to severe hypodysplasia or renal agenesis, resulting from failure of the UB formation or branching morphogenesis [5]. *Pax2*, a paired box gene, is expressed from E8.5 in the intermediate mesoderm and in the MM, WD and later in the UB. It is required to interpret the inductive signals coming from the UB, and regulates *Gdnf* transcription *in vitro* [6].

The signals that specify intermediate mesoderm have received more attention and been classified into two groups. *Eya1*, *Six1*, *Wt1* and *Hox11* specify the intermediate mesoderm along the anterior-posterior axis. In contrary, *Pax2/8*, *Lim1* and *Odd1* specify the intermediate mesoderm along the mediolateral axis [1,3]. Remarkably, *Six1-Eya* axis is critical regulator of both nephrogenic cord progenitors and the development of nephric duct [7]. In addition, the interactions between *Eya1* and both *Myc* and *Six2* transcription factors is critical for the expansion of the nephric progenitor pool during kidney development [8].

Eyes absent 1 (*Eya1*) encodes a transcriptional co-activator for *Six* genes and is essential for the MM specification [3,9]. *Eya1*$^{-/-}$ mutant mice show a combination of branchial, otic and renal anomalies, whereas the Wilms tumour suppressor gene, *Wt1*, is expressed exclusively in the MM and is necessary for *Gdnf* expression and survival of the MM-uninduced cells [3,9]. Absence of *Sall*, another MM-derived gene, results in incomplete UB outgrowth and failure of tubule formation [3,9,10]. UB outgrowth is perturbed and the MM does not express *Gdnf* when all sex alleles of the HoxII paralogous are deleted, with no change in *Eya1* and *Pax2* expression [11]. *Pax2* and *Pax8* are co-expressed in the WD, pro-and mesonephrons and have a redundant role in kidney lineage commitment. In *Pax2*-deficient mice, both the UB and kidney fail to form; however, the MM is morphologically distinct. *Pax2*/*Pax8* compound mutant mice fail to form pro- or mesonephrons [3,9]. *Lim1* is expressed in the WD, pro-and mesonephrons, and required for the correct patterning of all the intermediate mesoderm-derived epithelial structures. *Lim1*$^{-/-}$ mice exhibit disorganised intermediate mesoderm and lack *Pax2* expression [3,9]. Another gene, the mammalian ortholog of Timeless (mTim), is also important for epithelial morphogenes during early stages of renal development [3,9]. In addition, Smad4 protein controls the differentiation of ureteric smooth muscle cells during embryogenesis [3,9], while *Tbx18* is a critical regulator of the development of both vasculature network and glomerular mesangium in the kidney [8].

The vertebrate *Six*-homeobox genes (*Six1*-*Six*6) are homologous of Drosophila sine oculis. *Six* genes are essential for compound eye formation, synergistically with eyeless (*Pax* orthologs), *Eya* and dachshund genes [9]. The *Six* proteins contain a unique *Six* domain and the *Six*-type homeodomain, both of which are essential for specific DNA-binding and for interactions with *Eya* proteins [9]. *Six1*$^{-/-}$ mouse neonates do not survive and show anomalies in many organs, including kidney, inner air and skeletal muscle [9]. In *Six1*-deficiency, the metanephric mesenchyme is smaller in size, the UB fails to branch and the expression of several genes is disrupted: *Gdnf* and *Pax2* expression is partially reduced and *Sall1* and *Six2* are absent [9,12]. In addition, *Six1* controls the expression of *Grem1* in the metanepheric mesenchyme that is critical to induce branching morphogenesis in the developing kidney [9].

More severe kidney phenotype has been reported in *Eya*1$^{-/-}$ mice, which lack metanephric mesenchyme and UB formation with no expression of *Gdnf* and *Pax2* [9]. This raises the possibility that other *Six* family gene(s) cooperate with *Eya*1 and rescue, at least partially, *Six1* absence during early renal development. [9]. One possibility is the involvement of *Six4* in early kidney development. *Six4*, a gene separated by 100 kb from *Six1* on the same chromosome, shows a remarkably similar and overlapping expression pattern to *Six1* in a variety of vertebrate embryonic tissues [13].

However, *Six4*$^{-/-}$ mouse presents and shows little anomaly in embryogenesis. This suggests that *Six1* and *Six4* have a redundant role during organogenesis, considering their similarity in expression pattern, in binding specificity to MEF3 site of the myogenin promoter, synergistically with *Eya1*, and in targeting several common genes [9,13]. Indeed, *Six1* and *Six4* synergistically control the early steps of myogenic cell delamination and migration from the somite by regulating Met and *Pax*3 expression, and regulate the morphogenetic movements of early thymus/parathyroid tissues [9,13].

Other signals, molecules and factors are also critical for early nephrogenesis. A group of enzymes called Histone deacetylases (HDACs) play important roles in the regulation of kidney development [14]. In addition, the scaffolding proteins Talin, which bind to and may activate integrin, are crucial for the development of collecting ducts in the kidney [15], while the transcription factor *Myc* cooperates with β-catenin to induce the progenitor renewal program in the developing kidney.

Several *Wnt* genes, including *Wnt11*, *Wnt2b*, *Wnt4* and *Wnt7b*, regulate cell proliferation and embryonic morphogenesis and have unique expression domains within the embryonic kidney [16]. *Wnt11*, for example, is uniquely expressed in the branching ureteric tips at all stages of ureteric development, and regulates ureteric branching, at least in part by regulating *Gdnf* expression [17]. *Wnt11* expression is correlated with the initiation of ureteric branching since blocking of ureteric branching associates with loss of *Wnt11* expression. Furthermore, ureteric *Wnt11* expression is reciprocally dependent upon Ret/*Gdnf* signaling where implantation of *Gdnf*-coated beads causes induction of ectopic ureteric tips and increase of *Wnt11* at these sites [17]. *Wnt2b* is uniquely expressed in renal stroma and stimulates ureteric growth and branching in culture [16], suggesting an inductive role for renal stroma in kidney organogenesis.

Members of the *FGF* family are expressed in the developing kidney [18]. *FGF7*and *FGF10* have overlapping expression pattern in renal mesenchyme, and are most strongly implicated in the UB branching morphogenesis through binding to the UB-expressed *FGFR2b* receptor [19,20]. Lacking either *FGF7*or *FGF10* results in slightly smaller kidney, with reduced nephron number in the case of *FGF7* [19,20]. A similarity in receptor binding properties and overlapping expression suggests that *FGF7*and *FGF10* is partially redundant for *in vivo* kidney development.

References

1. Little MH, McMahon AP. Mammalian kidney development:principles, progress, and projections.Cold Spring Harb Perspect Biol. 2012;4(5).
2. Cullen-McEwen LA, Caruana G, Bertram JF. The where, what and why of the developing renal stroma. Nephron Exp Nephrol. 2005;99:E1-E8.
3. Xiao Q, Rongfei W, Lingqiang Z, et al. The roles of signaling pathways in regulating kidney development. Yi Chuan. 2015;37(1):1-7.
4. Durbec P, Marcos-Gutierrez CV, Kilkenny C, et al. GDNF signaling through the Ret receptor tyrosine kinase. Nature. 1996;381:789-93.
5. Moore MW, Klein RD, Farinas I et al. Renal and neuronal abnormalities in mice lacking GDNF. Nature. 1996;382:76-9.
6. Brophy PD, Ostrom L, Lang KM, et al. Regulation of ureteric bud outgrowth by Pax2-dependent activation of the glial derived neurotrophic factor gene. Development. 2001;128:4747-56.
7. Xu J, Xu PX. Eya-six are necessary for survival of nephrogenic cord progenitors and inducing nephric duct development before ureteric bud formation. Dev Dyn. 2015;244(7):866-73.
8. Xu J, Nie X, Cai X, et al. Tbx18 is essential for normal development of vasculature network and glomerular mesangium in the mammalian kidney. Dev Biol. 2014;391(1):17-31.
9. Xu PX. The EYA-SO/SIX complex in development and disease. Pediatr Nephrol. 2013;28(6):843-54.
10. Nishinakamura R, Matsumoto Y, Nakao K, et al. Murine homolog of SALL1 is essential for ureteric bud invasion in kidney development. Development. 2001;128:3105-15.
11. Wellik DM, Hawkes PJ, Capecchi MR. Hox11 paralogous genes are essential for metanephric kidney induction. Genes Dev. 2002;16:1423-32.
12. Xu PX, Zheng W, Huang L, et al. Six1 is required for the early organogenesis of mammalian kidney. Development. 2003;130:3085-94.
13. Kawakami K, Sato S, Ozaki H, et al. Six family genes-structure and function as transcription factors and their roles in development. Bioessays. 2000;22(7):616-26.
14. Chen S, El-Dahr SS. Histone deacetylases in kidney development:implications for disease and therapy. Pediatr Nephrol. 2013;28(5):689-98.
15. Mathew S, Palamuttam RJ, Mernaugh G, et al. Talin regulates integrin β1-dependent and independent cell functions in ureteric bud development. Development. 2017;144(22):4148-58.
16. Lin Y, Liu A, Zhang S, et al. Induction of ureter branching as a response to Wnt-2b signaling during early kidney organogenesis. Dev Dyn. 2001;222(1):26-39.
17. Majumdar A, Vainio S, Kispert A, et al. Wnt11 and Ret/Gdnf pathways cooperate in regulating ureteric branching during metanephric kidney development. Development. 2003;130(14):3175-85.
18. Cancilla B, Davies A, Cauchi JA, et al. Fibroblast growth factor receptors and their ligands in the adult rat kidney. Kidney Int. 2001;60(1):147-55.
19. Qiao J, Uzzo R, Obara-Ishihara T, et al. FGF-7 modulates ureteric bud growth and nephron number in the developing kidney. Development. 1999;126(3):547-54.

20. Ohuchi H, Hori Y, Yamasaki M, et al. FGF10 acts as a major ligand for FGF receptor 2 IIIb in mouse multi-organ development. Biochem Biophys Res Commun. 2000;277(3):643-49.

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