

Urinary Proteomic Analysis at Termination of Nephrogenesis in Mice

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The kidney develops through reciprocal interactions between ureteric bud tips and surrounding mesenchyme. The interaction produces new nephrons and continues for 3 days after birth in mice. The mechanism triggering the end of nephrogenesis is unknown, but occurs when there are multiple birth-related physiologic changes including increases in renal blood flow and glomerular filtration. One hypothesis is that these physiologic changes initiate the end of nephrogenesis by increasing the excretion of growth factors. We performed urinary proteomic analysis during the critical postnatal time. Urine proteins from timed collections were separated by PAGE. Differently excreted proteins were analyzed by MALDI-MS. Western blot analysis was used to examine candidate growth factors and to validate spectrometry results. We found that the urinary protein excretion profile changed immediately after birth. Albumin excretion was constant indicating stable glomerular permeability in the first 24 hours after birth and suggesting that the changes in urine proteins must be due to alterations in plasma proteins or in tubular function. Western blot analysis of candidate genes, *Sfrp1*, *Bmp7* and *Fgf8*, showed no change during the first 48 hours after birth. Mass spectrometry analysis of differently excreted proteins showed increases in uromodulin, meprin A, complement factors B and D, *Ltbp4*, and *Tff3*. Microarray databases showed the source of uromodulin and meprin to be the distal and proximal tubule, respectively. Other changes were probably due to increased plasma concentration. Examining the end of nephrogenesis is important because it ultimately controls final nephron number. We did not detect changes in critical growth factors. This could occur either because of insensitivity or insignificant growth factor excretion. Future studies will use affinity matrices to purify urinary proteins.