

Figure S1. Methodology for IMC optimization and downstream analysis.



Figure S2. Validation of immune markers on human lymph node and skin abscess tissue. Markers and tissue type are indicated. **(A)** In lymph node, CD20-positive B-cells are located in germinal centers (GC), with CD3-positive T-cells interspersed. T-cells are subdivided into CD4- and CD8-positive populations. CD68-positive macrophages are found throughout the tissue but excluded from sinusoids (Sin). **(B)** In abscess tissue, CD66b and CD68 expression are mutually exclusive. Scale bars: 100µm.



Figure S3. Transitional areas identified by IMC. (A-D) IMC reconstructions of a selected region showing distal convoluted tubule (DCT) transitioning into connecting tubule (T). Scale bar (A) 100µm.



Figure S4. Identification of infiltrating cells in human kidney with IMC. (A-G) Individual images from the same region of a nephrectomized kidney, false colored for each marker as indicated. **(H)** Merged image from all seven channels from (A-G). Arrowheads in (G and H) show a CD66b-CD68-double-positive cell. Scale bar: 100µm (A).



Figure S5. Reproducibility of *Kidney-MAPPS* (A) H&E stained region of tumor remote nephrectomy tissue. Insets show the regions selected for the 2 separate IMC imaging ablations, shown in detail in (B) and (C). (B-C) Pseudocolored IMC images with channels indicated. Insets show regions defined morphologically as cortex and selected for quantitative analysis for validation purposes. (D) Squares represent quantitative data of cellular proportions from original regions of interest ablated and analyzed. Triangles represent paired regions from the same kidney section, stained at the same time, and analyzed separately. Bars represent mean values. N = 4 pairs. (E) Squares represent quantitative data from the original regions analyzed in D. Triangles represent data from the same regions imaged on adjacent sections, with staining performed several months later using a new antibody cocktail. Bars represent mean values. N = 4 pairs. The Wilcoxon matched-pairs signed ranked test was used to determine statistical significance, with p values indicated. Glom – glomerular. Endo (I) – interstitial endothelium. Endo (G) – glomerular endothelium. Scale bars 5mm (A) and 600µm (B).



Figure S6. Defining inner and outer stripe of the renal medulla. (A) Proportions of tubular cell types from each of nine medullas from normal kidneys, ranked in descending order of proximal tubule proportion. **(B)** The distance of each medullary region's midpoint graphed against the proportion of total proximal tubule detected in each sample.

Table S1. Characteristics of nephrectomy patients and living donors

N	Nephrectomy						
A	Age	Female Sex	Serum creatinine (mg/dL)	Estimated GFR	Hypertension?	Diabetes?	Pathology
6	62	Ν	1.2	>60	Ν	Ν	Non-neoplastic kidney shows no significant pathological changes.
6	68	Y	0.5	>60	Y	Ν	Non-neoplastic kidney shows no significant ultrastructural abnormalities
5	54	Ν	1	>60	Ν	Ν	Non-neoplastic kidney shows no significant pathological changes
4	48	Y	0.82	>60	Y	Ν	Uninvolved kidney, normal parenchyma
3	31	Ν	unknown	unknown	unknown	unknown	Uninvolved kidney without significant abnormalities
5	56	Ν	0.94	>60	Ν	Ν	Non-neoplastic kidney unremarkable
6	69	Ν	1.2	60	Y	Ν	Uninvolved kidney parenchyma is unremarkable
5	53	Ν	unknown	>60	unknown	unknown	Uninvolved kidney without significant abnormalities
5	51	Y	0.69	>60	Ν	Ν	Non-neoplastic kidney shows minimal histopathologic changes
5	52	Ν	0.87	>60	Y	Ν	Non-neoplastic kidney shows minimal histopathologic changes
6	64	Y	0.81	>60	Ν	Ν	Non-neoplastic kidney shows minimal histopathologic changes
5	55.3	4/11	0.89	9/9	4/9	0/9	
L	Living Donor						
4	41	Y	0.75	>60	Ν	N	No significant histopathological changes
2	29	Y	0.76	>60	Ν	Ν	No significant histopathological changes
3	37	Ν	1.05	>60	Ν	Ν	Mild interstitial fibrosis < 5%
6	64	Y	0.78	>60	Ν	Ν	Mild interstitial fibrosis < 5%
6	62	Y	1	>60	Ν	N	Mild interstitial fibrosis < 5%
4	46.6	4/5	0.87	5/5	0/5	0/5	
P value 0	0.21	0.28	0.83	1.0	0.22	1.0	