KIDNEY RESEARCH INSTITUTE

Transforming Lives through Innovation and Discovery

Innovation in Kidney Disease Research: the Human Kidney on a Chip

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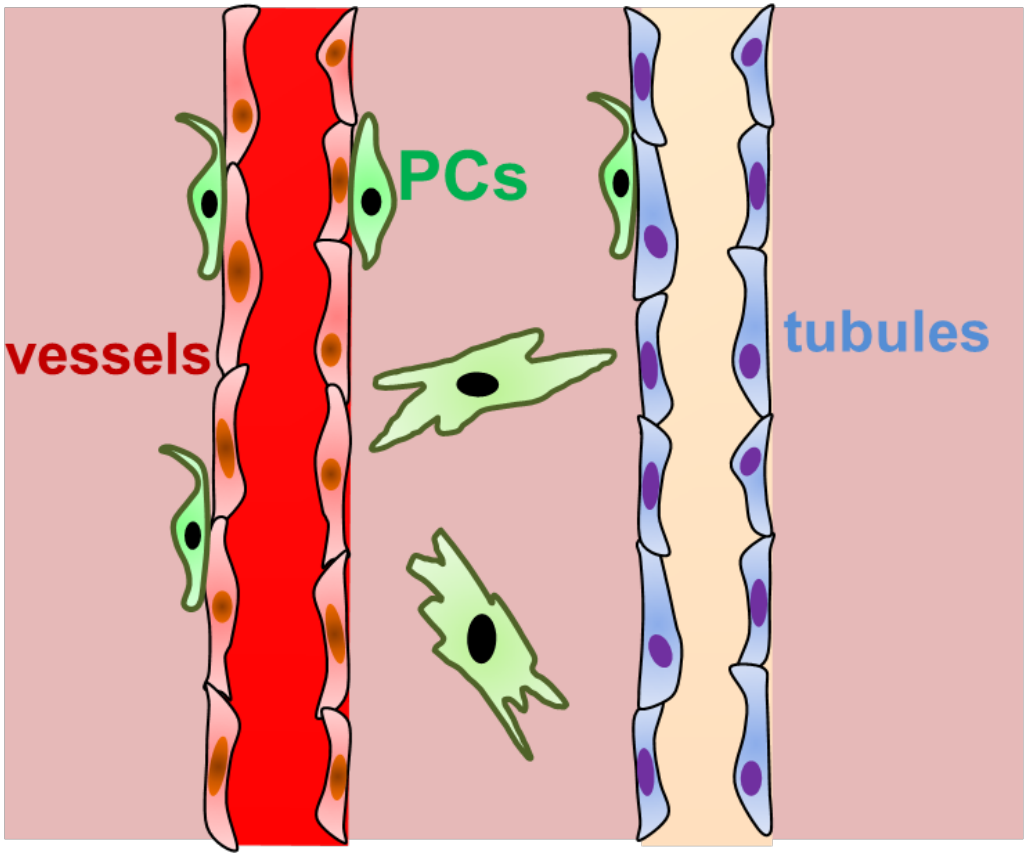
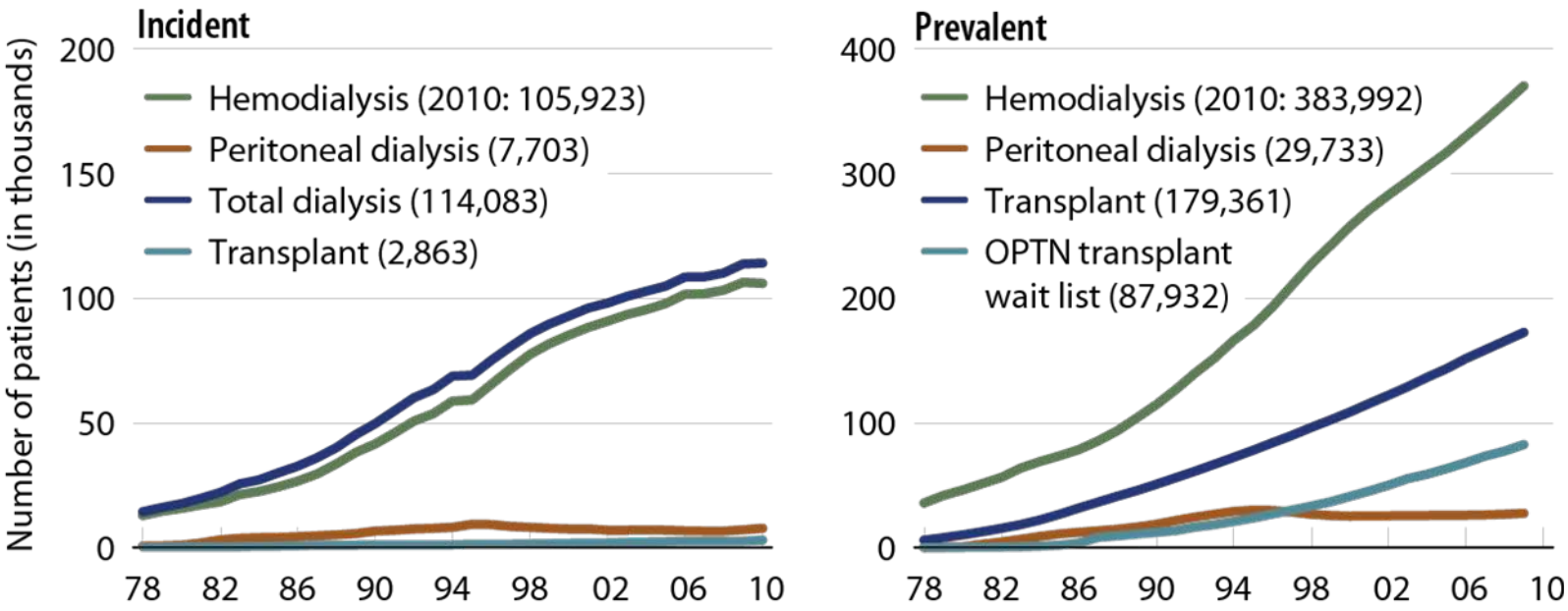
Why a Kidney on a Chip?

- Rationale
- Goals
- Approach
- Technology
- Progress to date
- Challenges
- Long term Plans



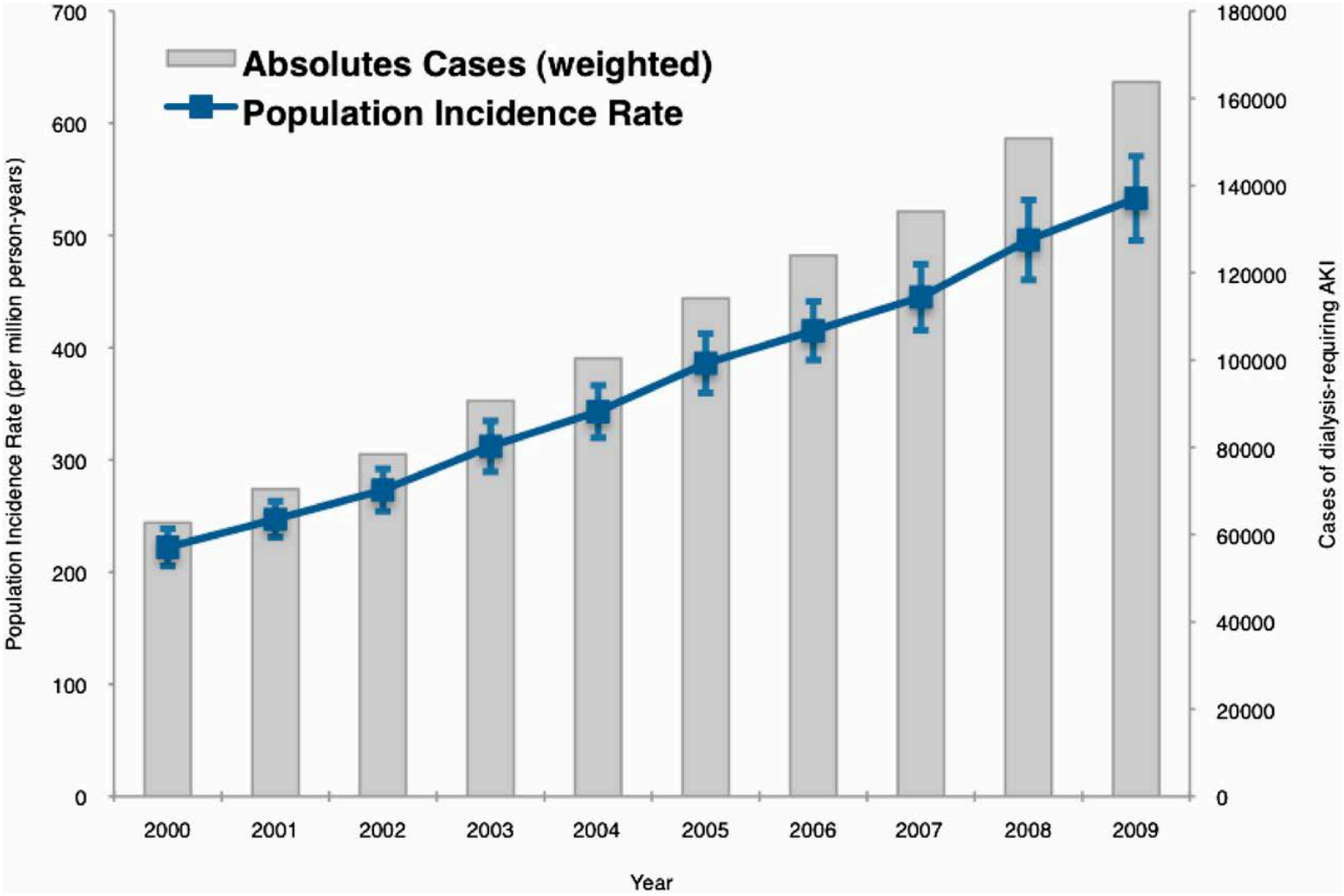
Unsolved Public Health Problems in Kidney Disease: 1960-2013



- Increasing incidence and prevalence of kidney disease
- Ongoing high morbidity, mortality and costs
- Lack of innovation in therapies
- Lack of high level evidence from clinical trials

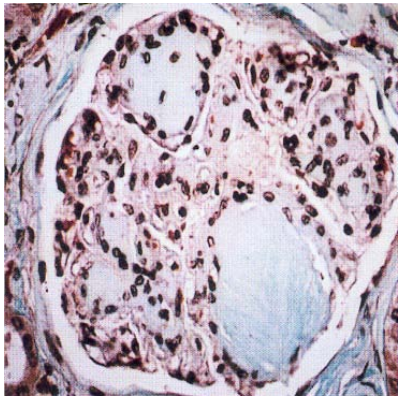
USRDS Annual Data Report 2012

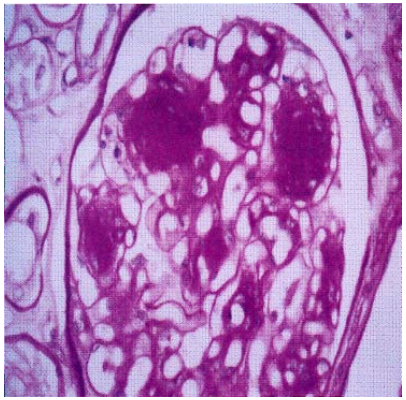
Population incidence of dialysis-requiring Acute Kidney Injury in the United States

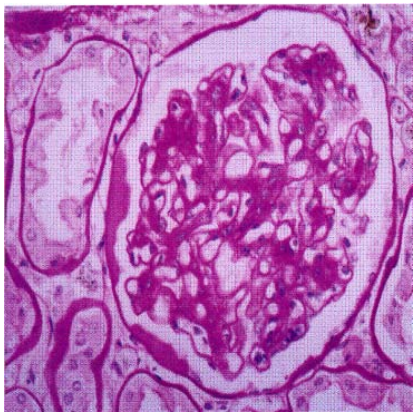


Hsu R K et al. JASN 2013;24:37-42

Epidemic of Diabetic Kidney Disease







- Incidence doubled in past decade
- Twelve fold increased risk of End-Stage Kidney Disease
- Leading cause of End-Stage Kidney Disease (54% of incident cases)
- Current treatment can slow disease progression
- No additional new therapies proven beneficial in past 10 years

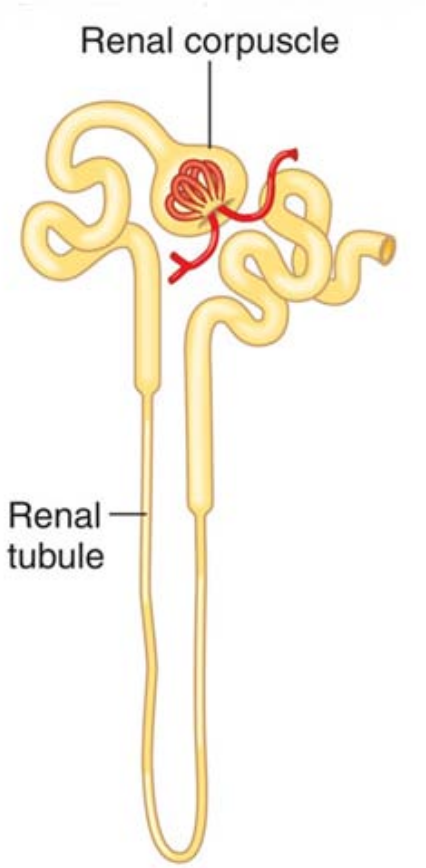
FDA APPROVED NEW MEDICAL ENTITIES (NME) FOR KIDNEY DISEASE IN THE LAST DECADE (2002 – 2012)

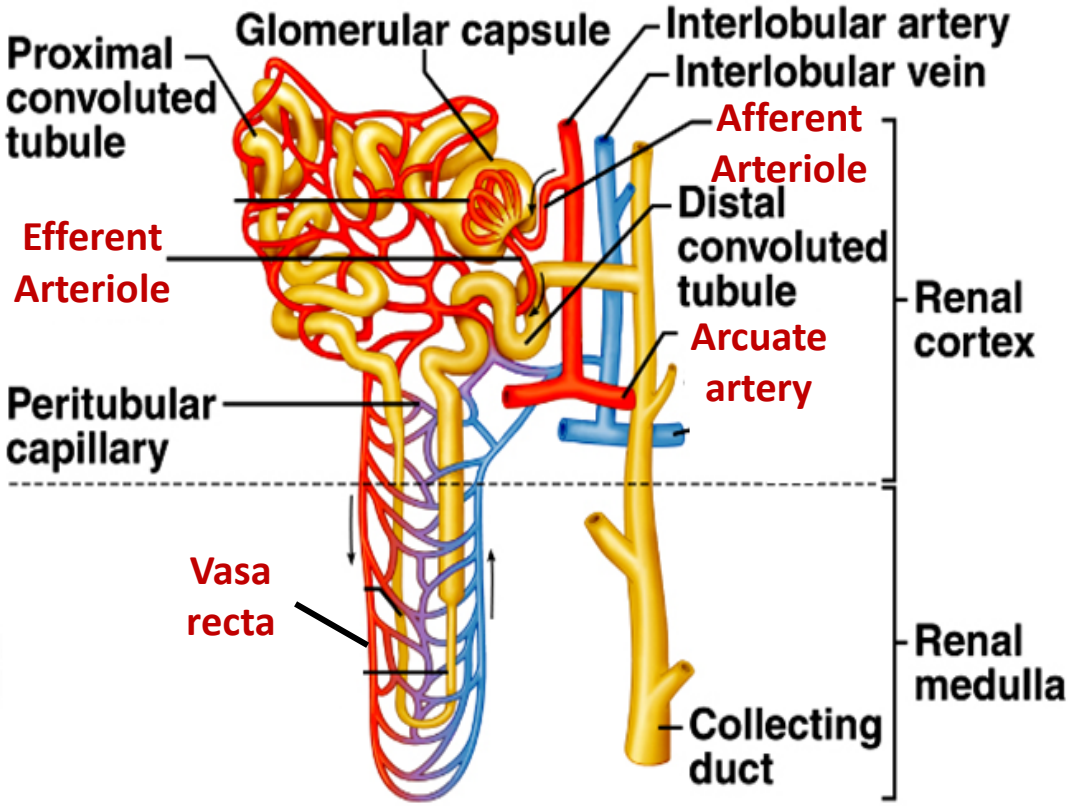
	Drug	Year	Indication
1.	Icodextrin	2002	Peritoneal Dialysate
2.	Cinacalcet	2004	Calcimimetic, Hyperparathyroidism
3.	Lanthanum	2004	Phosphorus binder, Hyperparathyroidism
4.	Convivaptan	2005	Vasopressin antagonist, Hyponatremia
5.	Mircera	2007	Erythropoietin stimulating agent, Anemia
6.	Tolvaptan	2009	Vasopressin antagonist, Hyponatremia
7.	Belatacept	2011	Immunosuppressive Drug, Transplantation
8.	Pegenesitide	2012	Erythropoietin stimulating agent, Anemia

Drug Therapy and Kidney Disease

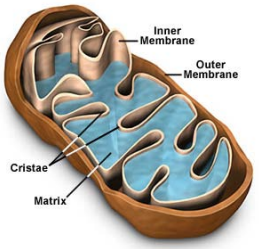
- Renal blood flow rate is 20-25% of cardiac output
- Kidney function plays a primary role in the elimination of 20-25% of drugs and their metabolites.
- The kidney is highly susceptible to injury from drugs.
- Over 20 million adult Americans have kidney disease, which alters drug metabolism and elimination.
- Up to 20% of all hospital admissions for community acquired acute kidney injury are attributable to drug induced kidney injury.
- People with kidney disease are at greatly increased risk of adverse drug reactions.

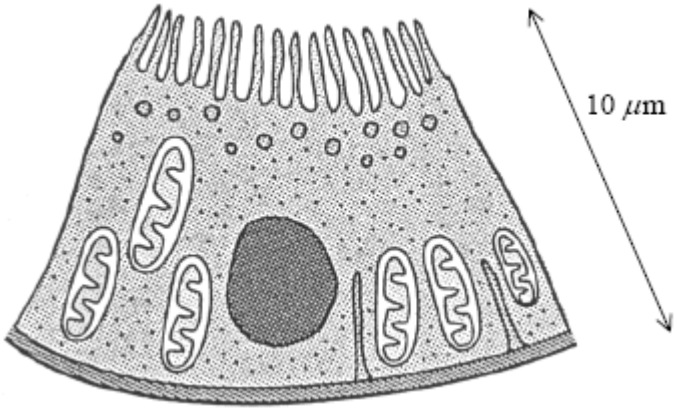
The Nephron: the functional unit of the kidney

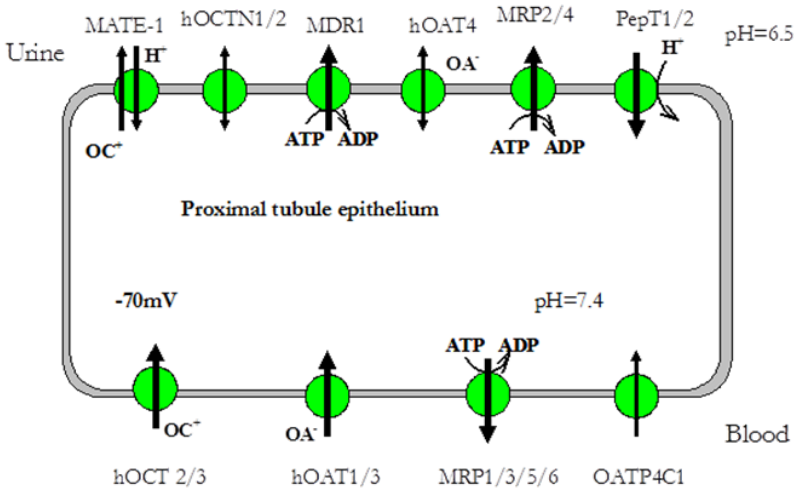




Proximal Tubule







- **Reabsorption**
  - Sodium (65%) and water (65%).
  - HCO_3 (80-90%)
  - Others: K (65%); Ca (80%); PO_4 (90%); Mg (20%); Glucose (100%); uric acid (90%)
- **Secretion**
  - Cations (Creatinine, drugs [e.g. trimethoprim, cimetidine])
  - Anions (hippurate; drugs [e.g. diuretics, penicillin, cephalosporins, salicylates])

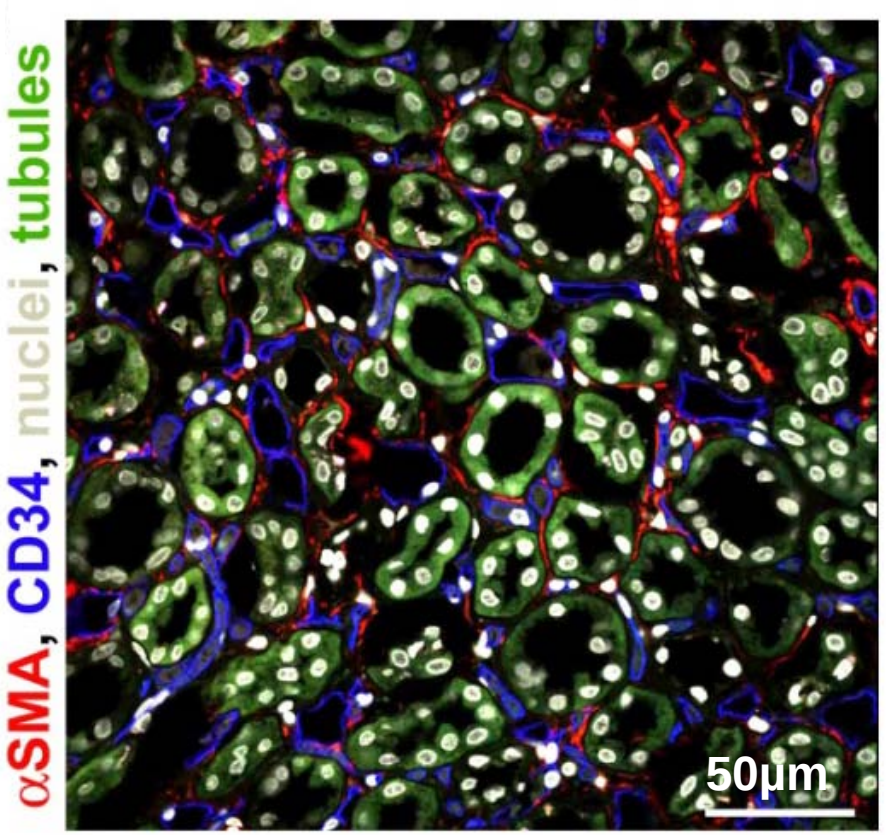
- **Ammoniagenesis**

generation of NH_4^+ from glutamine which is secreted into lumen by sodium hydrogen antiporter (NHE3)

- **1α-hydroxylation** of 25(OH)Vit D to form 1,25 $(OH)_2$-Vit D (calcitriol).

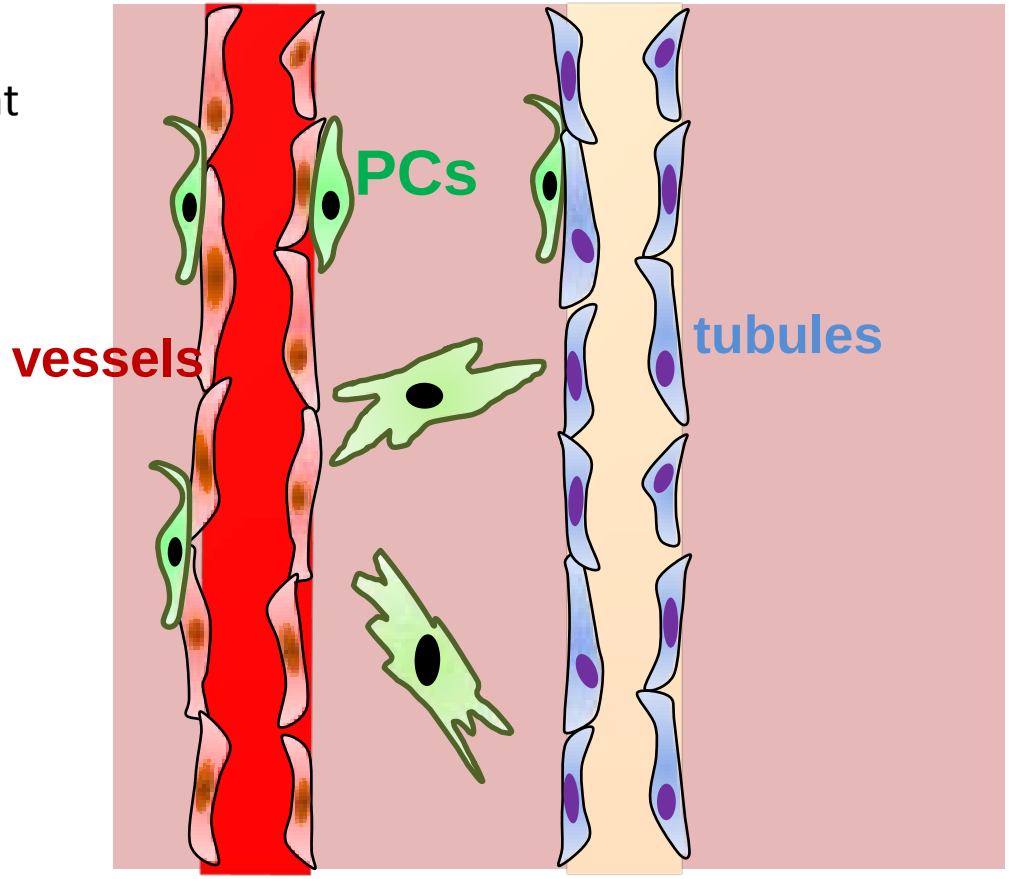
Kidney Drug and Toxin Clearance

$$CL_R = \frac{Excretion\ rate}{Plasma\ conc} = (1 - Frac\ Reabs)\left[\frac{Filtration\ rate}{Plasma\ Conc} + \frac{Secretion\ rate}{Plasma\ conc}\right]$$



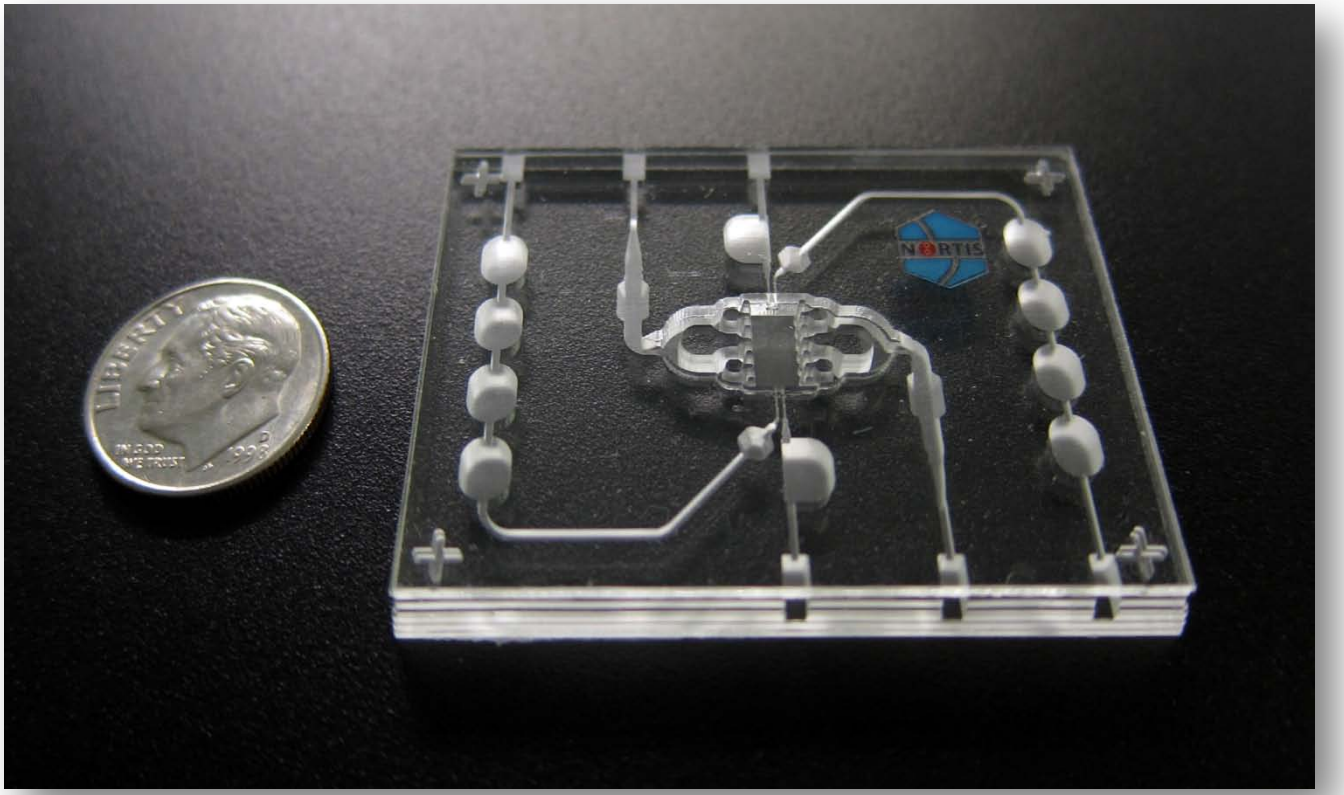
Kidney Tubulo-Interstitium on a Chip

- The primary goal is to design, implement and test a tissue engineered human kidney microphysiological system.
- The system will be developed to fully evaluate uptake, metabolism and elimination of xenobiotics in a human tissue derived, in vitro 3-dimensional system that accurately reflects human physiology.
- The microphysiological system can be used to assess the response to organ injury inflicted by endogenous and exogenous toxicants.



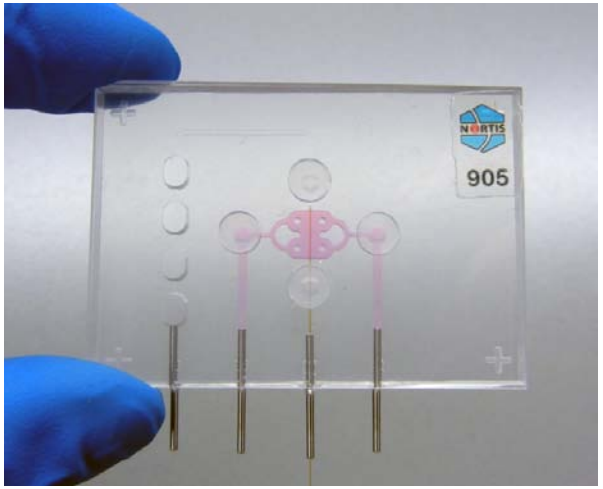
Nortis Chip Technology

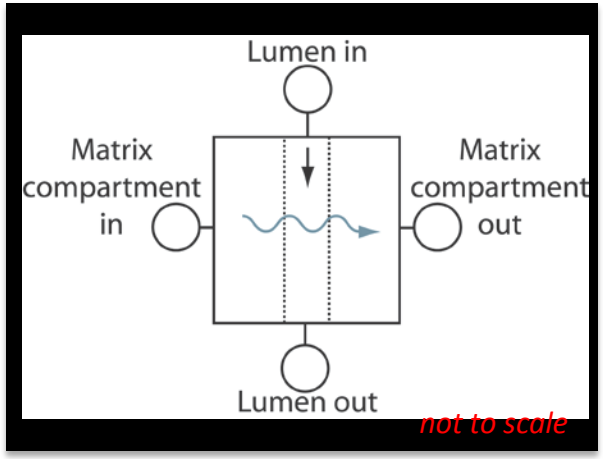
Disposable, chip-like devices for the creation of vascularized 3D microenvironments of human tissues/organs



Nortis 3-D Cell Culture Chip Technology

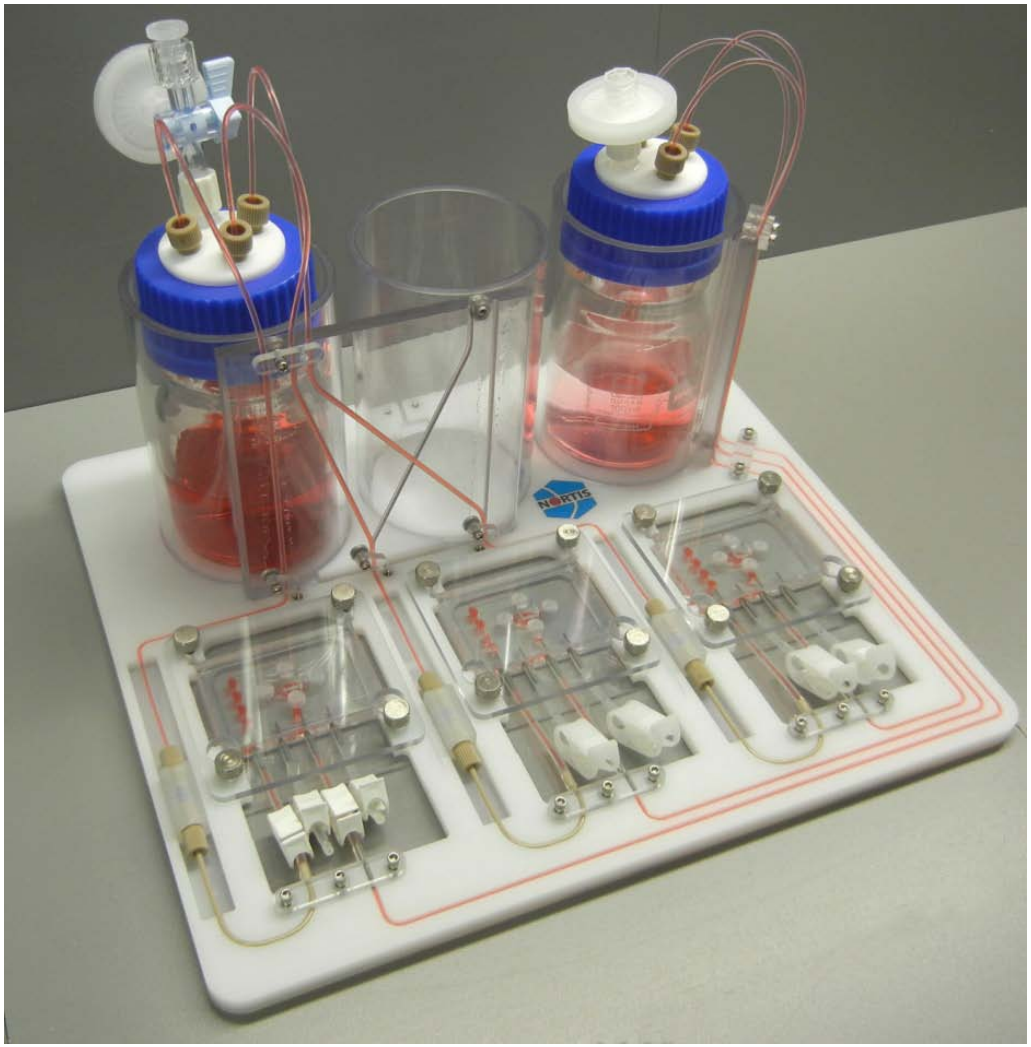
- Disposable microfluidic chips containing 3D micro-environments that are traversed by one or more tubular cell structures
- The setup allows for creation of compartmentalized tissue models: lumenal versus extracellular matrix (ECM) compartment
- Both, lumenal and ECM compartments can be independently perfused
- There are no artificial material surfaces to which cells must attach
- Lumenal fluid flow leads to controlled shear force and other mechanical stimuli
- Septa allow for injection/extraction of fluids directly on the chip, and insertion of sensors
- Integrated bubble-traps





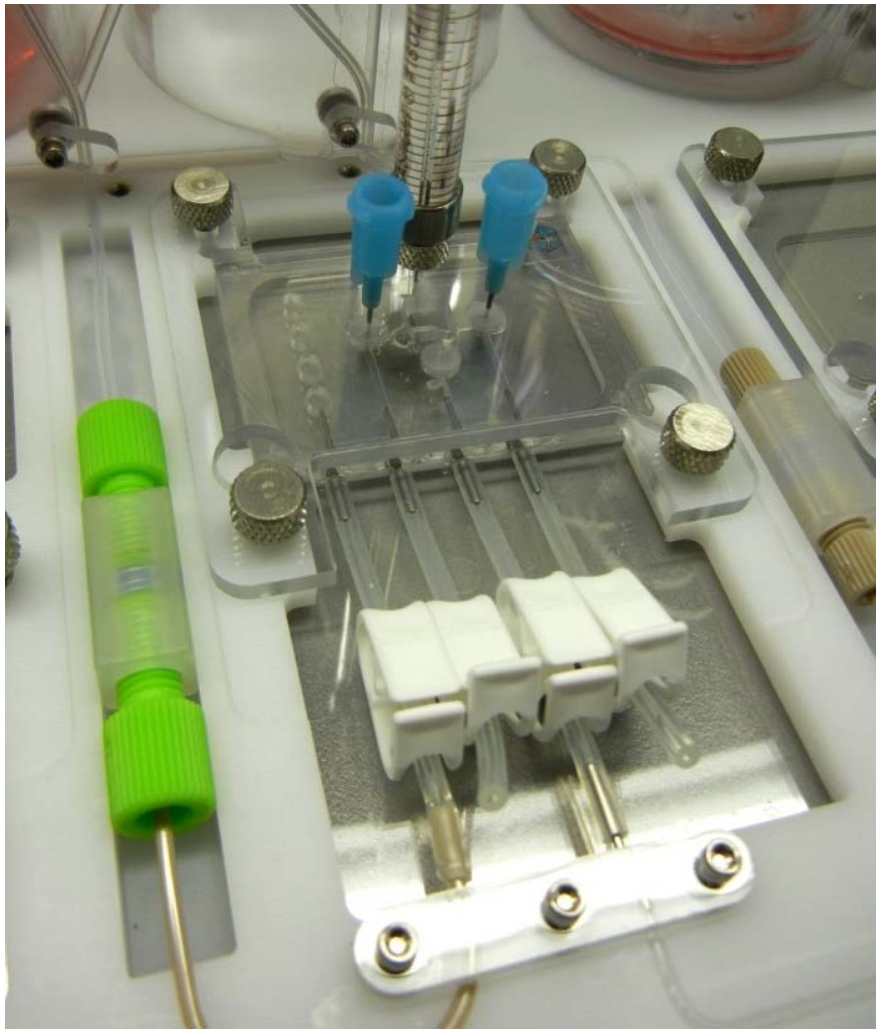
Platform for Perfusion and Environmental Control

- Designed to be used in a standard cell-incubator
- Fluid control accomplished using pressurized air (same composition as desired in cell incubator) to perfuse fluids through cell culture devices
- Many platforms (dozens of microfluidic chips) can be pneumatically powered via one common pressurized gas connection
- Flow rates can be varied by changing flow resistor, or by connecting a separate pressure regulator
- High flow resistor value means flow rate more consistent with changing biological system fluidic resistance or other variations
- Quick disconnects from pressurized gas allow the platform to be easily and safely moved from incubator to microscope
- Self-contained platform means system is insensitive to fluidic perturbation (unlike when liquid supply lines are run from an external pump to a microfluidic device)

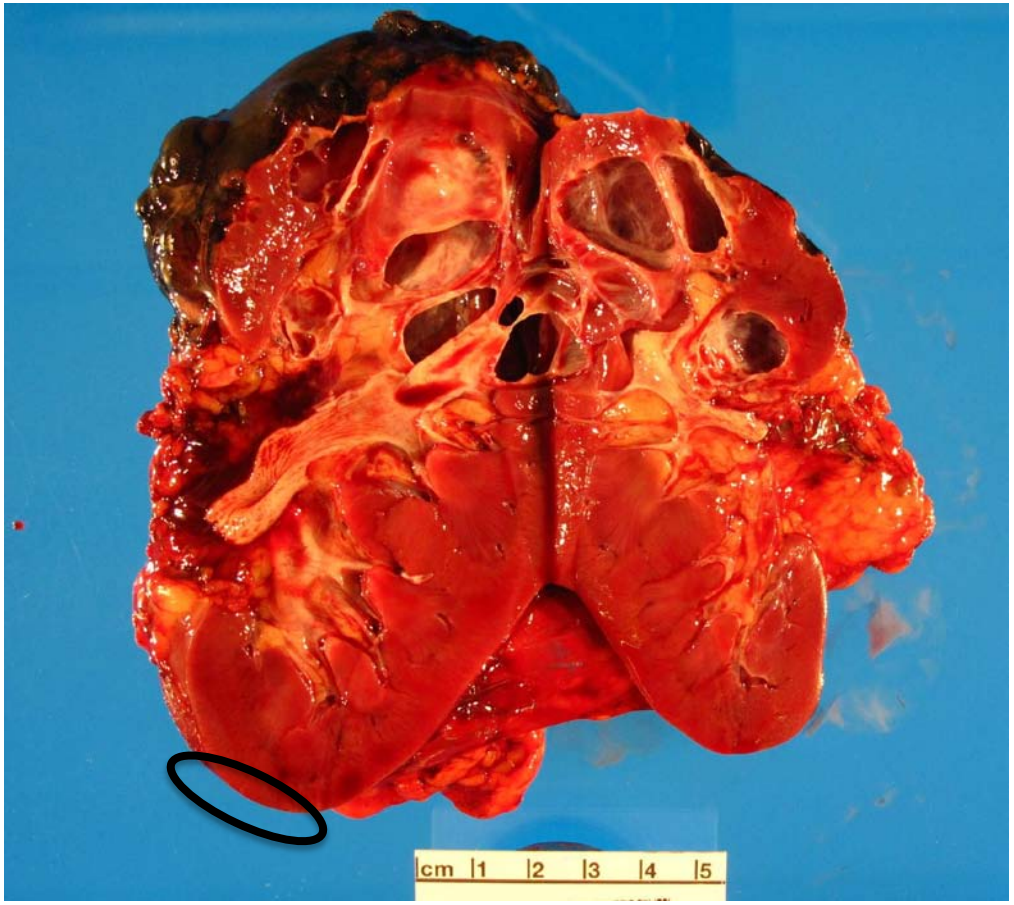


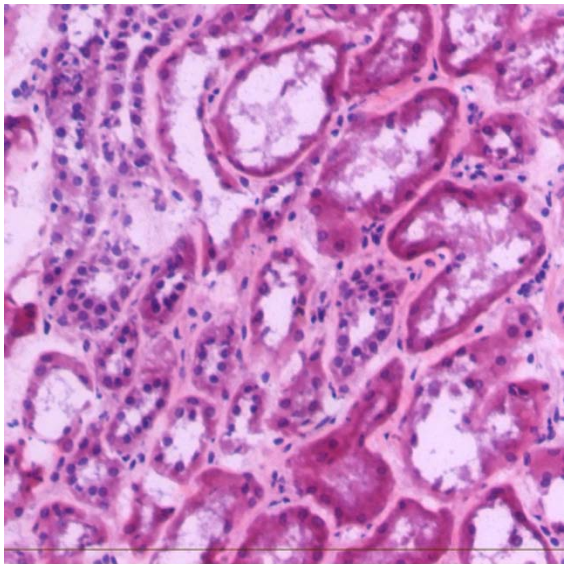
Platform for Perfusion and Environmental Control

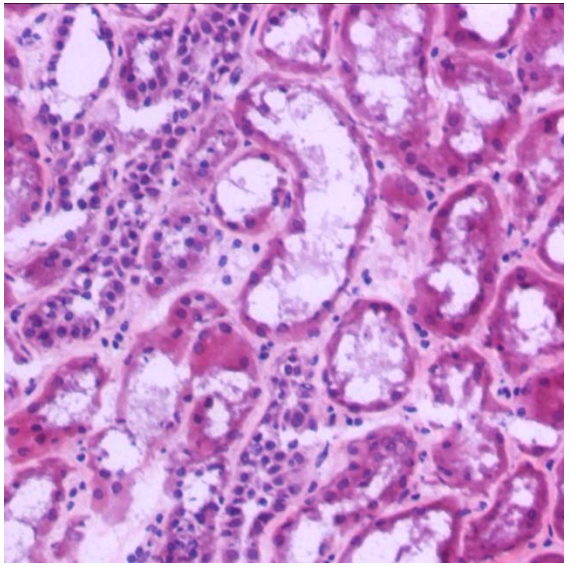
- Low-cost polycarbonate assembly provides tubing stabilization, mechanical support for pressure bottles, and cell culture device(s)
- Microscope-imaging friendly: Can image entire vessel in brightfield, phase contrast, fluorescence. Usable with objectives down to 0.65 mm working distance
- Autoclavable, easily cleaned
- Can be operated for long term experiments without much attendance (in contrast to other systems like gravity-feed perfusion, which require daily refilling of perfusate)
- Closed system: no problems with sterility or evaporation
- Can get 20x range of flow rates, probably higher. Typical range is 0.5 microliters/min to 10 microliters/min
- Needle guides on platform to regulate position and depth of needle injection
- Design-in-progress integrates pressure container into stage, eliminating large pressure bottles and excess swept volume. Makes use of 5mL Eppendorf tubes



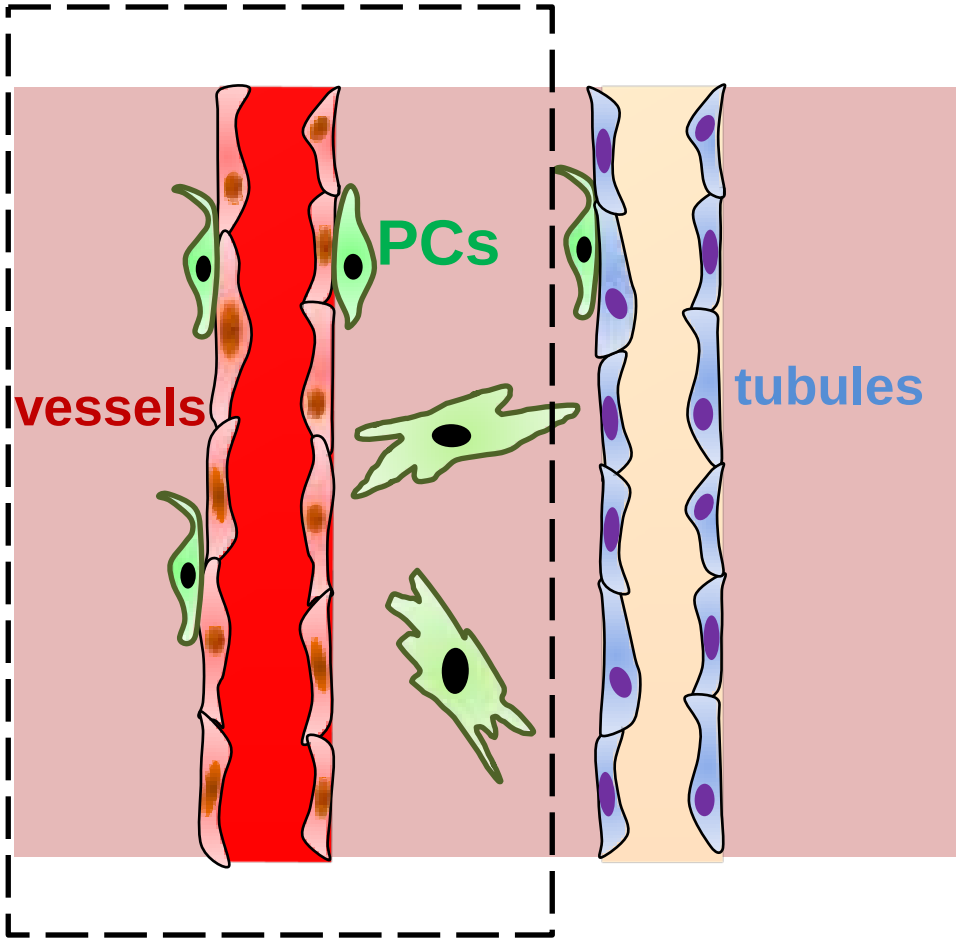
Cell sources: Isolation and purification of primary cells from normal human kidney







Kidney microvessels

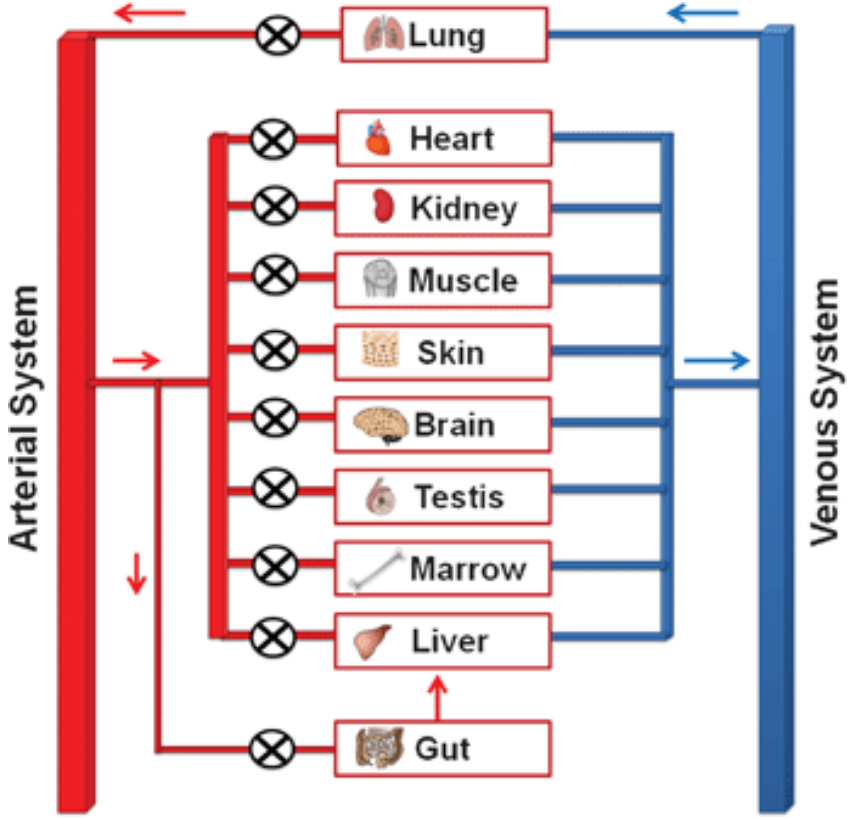


1. **Cell sources:**
   - **Peritubular endothelial cells**
     - Isolation – $CD34^{+}$
     - Characterization: morphology, proliferation, marker expression
   - **Perivascular cells**
     - Isolation – PDGFRb, NG2, CD73
     - Characterization: morphology, marker expression
2. **Microvessel formation**
   - Survivability
   - Permeability
   - Endothelial cell junctions
   - EC-PC interactions
3. **Microvessel – peritubules**

Short Term Challenges to System Optimization

- Cell source
- Cell seeding and adhesion
- Extracellular matrix
- Kidney derived microvessels
- Flow dynamics
- Analytical chemistry

An integrated multi-organ systems instrument



Huh et al, Lab Chip,2012,12:2156-2164

Longer term plans and goals: three to five year vision

- Merging of ‘top down’ and ‘bottom up’ approaches
- Consistent cell sources (cell lines?, iPSCs?)
- Consistent high quality real time biomarker and imaging readouts
- Enhanced ADMET prediction
- Genetic analysis of transporters and drug metabolizing enzymes (personalized medicine)
- “Offer the ability to produce high quality/high content data at key stages of the drug discovery process” (Ingber group)

Potential Additional Uses of the Kidney on a Chip

- Improved drug dosing in kidney disease
- Tool for understanding uremia
- Tool for improving organ preservation for kidney transplantation
- Tool for drug development for kidney disease
- Step towards an implantable artificial kidney?