Kidney-On-A-Chip

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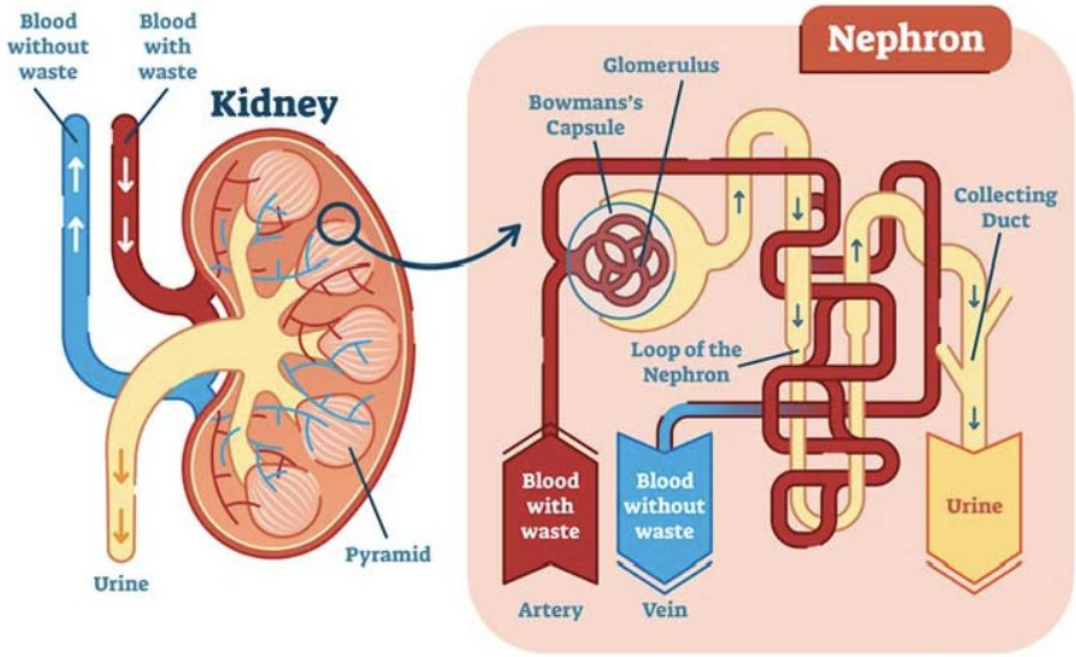
Background

Kidney Function

Blood filtration!

- Kidneys filter about 200 quarts of blood each day
- Rid the body of unnecessary materials and toxic waste

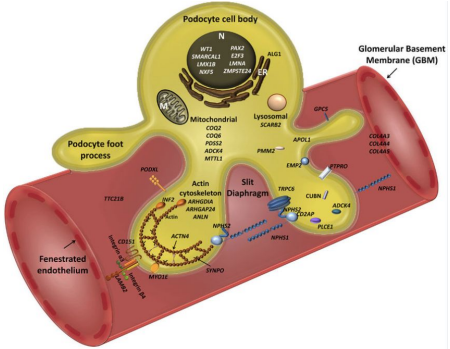
*The kidneys do so much more than just blood filtration, but this will be the focus



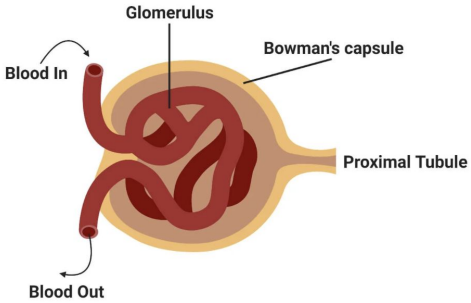
(Apollo Medics, 2024)

Important Elements

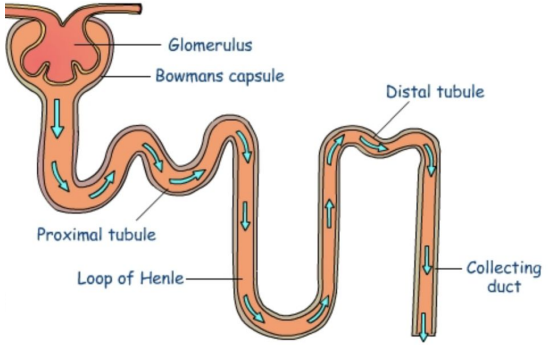
Podocyte



Glomerulus



Tubule



(Bierzynska, 2014; Dewangan, 2023; Toppr)

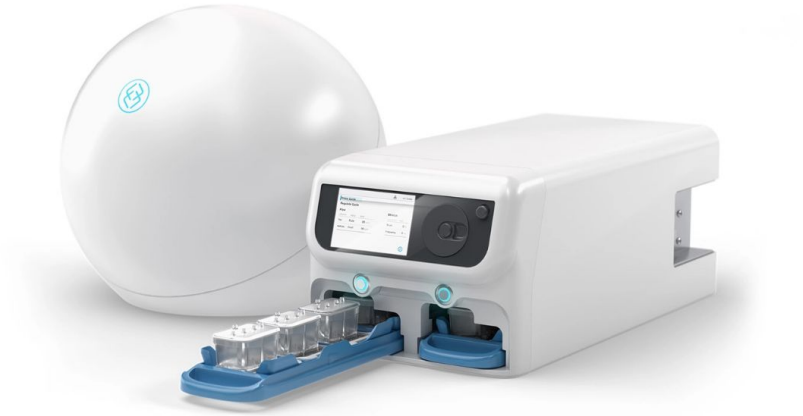
Kidney-On-A-Chip (KoC)

- First KoC developed in 2008
- KoCs have been used to study a variety of parameters, including flow, shear stress, and even organ–organ interactions in multi-organ-on-a-chip (MOC) models
- 2D cell systems have been replicated well in chip models, more advanced models are still mostly in development

(Kim et al, 2015)

Current Technology

- KoC vs glomerulus-on-a-chip, tubule-on-a-chip
- Emulate Kidney-Chip toxicology screening system



(Emulate Bio)

Applications

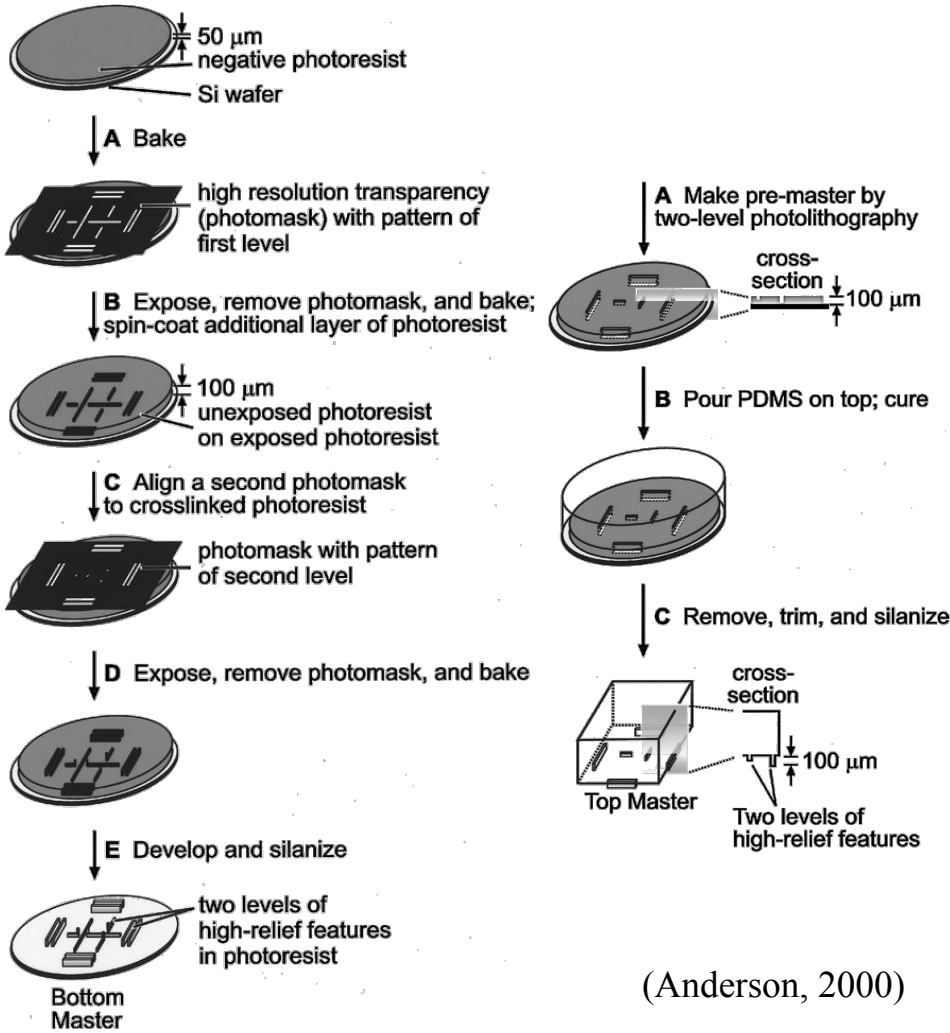
- Disease state modeling
  - Kidney diseases affect ~10% of the world population
- Studying renal physiology
- Tissue engineering
- Big possibilities in the pharmaceutical industry and testing applications for new drugs
  - As of 2023, the FDA no longer requires all drugs to go through animal testing – this could be a great alternative
- In 2019, UW sent a KoC to space to study how weightlessness alters kidney function

Fabrication

Microfabrication

The First KoC model was published in 2008:

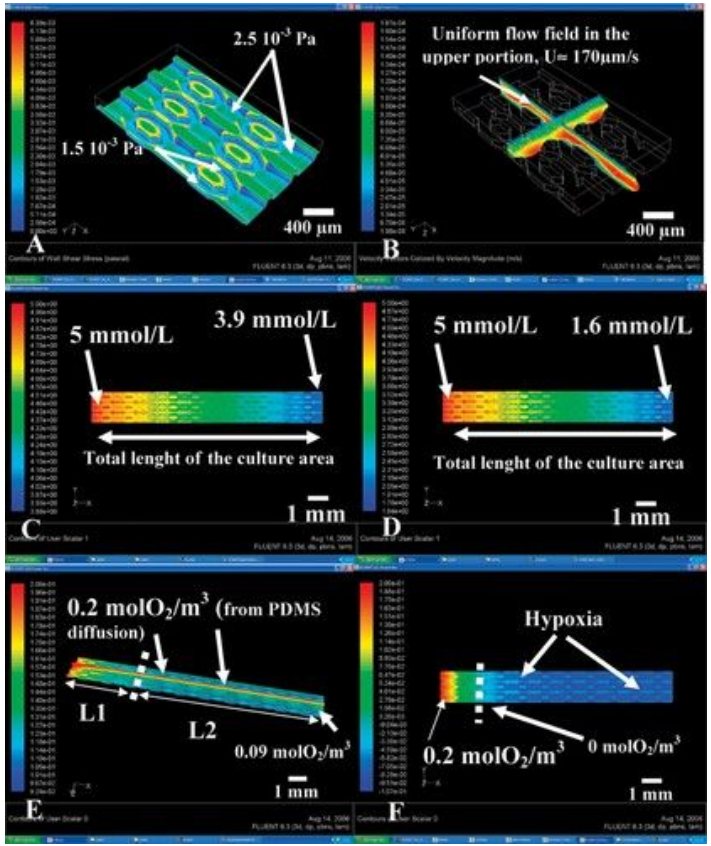
- Utilized lithography techniques to create a master mold
  - Poured PDMS on top and cured it to make the microfluidic platform
- Fibronectin coated on PDMS
- Chip had microchambers, microchannels and a larger cell culture chamber
  - Total volume of 30 μL with 2 cm^2 of surface area for cell growth



(Anderson, 2000)

Microfluidics

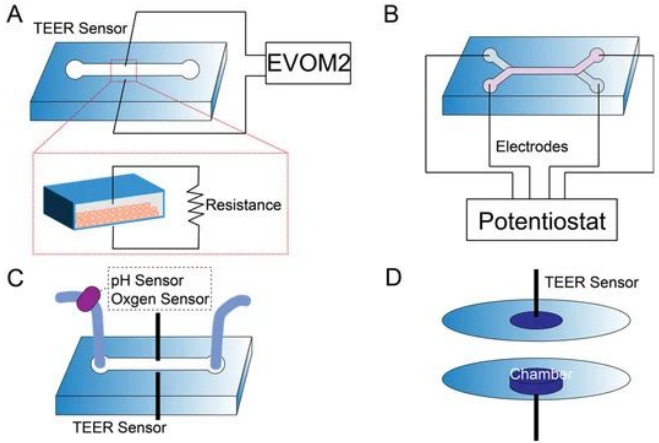
- Renal Cells were cultured to a confluence of 5×10^5 cells/cm^2 in Minimum Essential Medium
- Seeded and then allowed to rest for 12 hours to attach to the surfaces
- Perfusion (10 µL/min) was started, and after 96 hours the cells formed into a 3D structure
- These preliminary models were tested by counting the number of adhered cells and to quantify their metabolic activity as compared to physiological Kidney Cells.



(Baudoin, 2008)

Biosensors

- Sensors can be integrated into KoC to measure physical, electrical, electrochemical, and optical data.
- TEER, specifically, is often used in KoC to monitor epithelial barrier formation
- Real-time monitoring allows for better understanding of the characteristics of the system (cell growth, metabolism, clearance rates, etc.)
- Biosensors are often incorporated directly into the PDMS chip- electrodes can be parallel to or intersecting the channels.



(Luo, 2022)

Drug-Testing Improvements

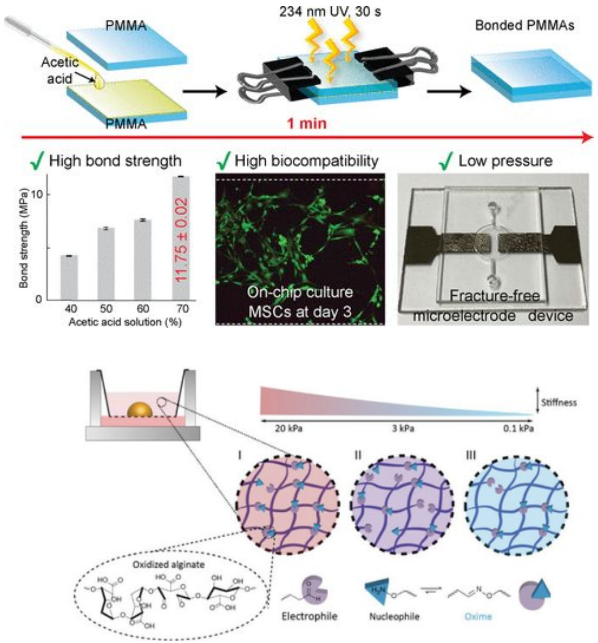
- Two current issues with KoC models:
  - Usage of PDMS allowing small-molecule absorption (inhibitory to drug screening)
  - Lack of vascularization and complexity in common models

- Proposition: implement PMMA instead of PDMS and create a more realistic ECM to encourage blood vessel formation, to ultimately make a better drug-screening model.

(Ashammakhi, 2018)

PMMA and Vascularization

- PMMA has low permeability for small molecules and mechanical properties compatible with organ-on-a-chip models.
  - Instead of soft lithography, PMMA can be engraved with laser micromachining and stacked in layers to form the desired architecture.
  - Bonding of these layers can be performed with acetic acid and UV irradiation at low pressure with clamps



- Vascularization can be supported via culturing in a hypoxic environment that mimics the physiologic kidney (7% O_2)
- The inclusion of a hydrogel coating for the cells to grow on can form a more physiological ECM.

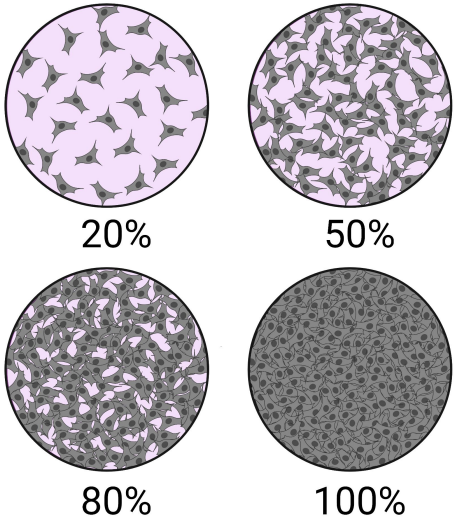
(Cao, 2023), (Loan Trinh, 2020), (Matellan, 2018), (Corral-Najera, 2023)

Testing & Validation

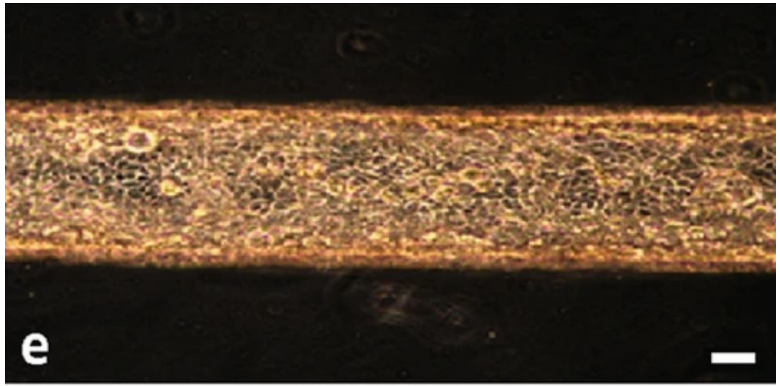
Structural Validation

First step of tissue engineering validation

- Checking that appropriate cells are present
- Understanding the general morphology
  - Epithelium formed correctly
  - Renal cells confluence
- Measuring cell viability and density
  - Biocompatibility testing



(Bestbier, 2024)

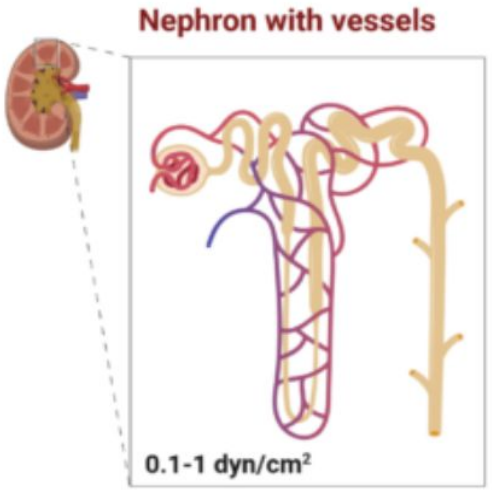


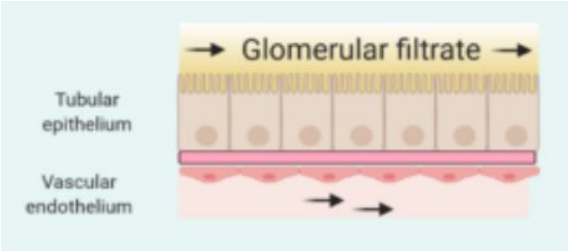
(Aceves, 2022)

Mechanical Testing

Mechanical stimulation influences cell differentiation and morphology

- PMMA/substrate stiffness
- Shear stress
  - From flow of blood in channels
- Pressure measurements
- Tensile/compression testing

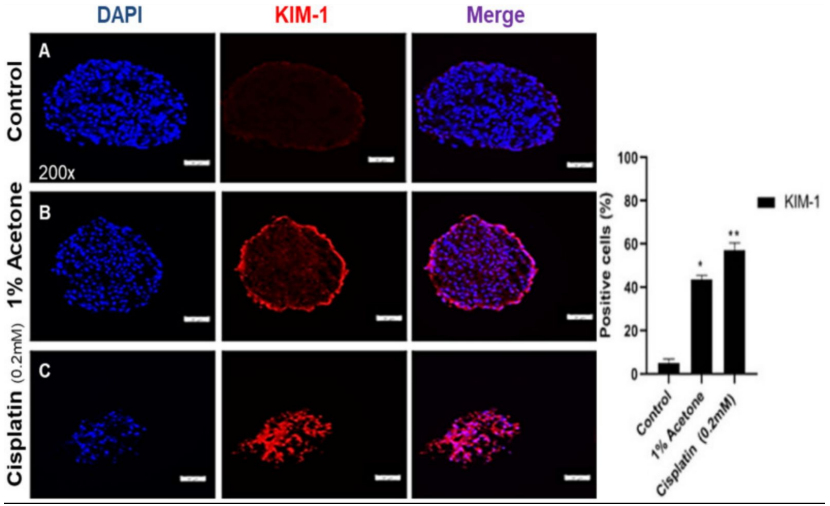




(BeOnChip, 2021)

Kidney Function Validation

- Metabolic activity
  - Testing the renal Cells
- Permeability
  - Ensure the PMMA is not absorbing proteins
- Filtering
  - Testing small molecules ability to cross the barrier
  - Tracking large proteins staying in the “plasma”
- Response to signals
  - Testing the response of the KoC in relation to natural cells



(Yu, 2023)

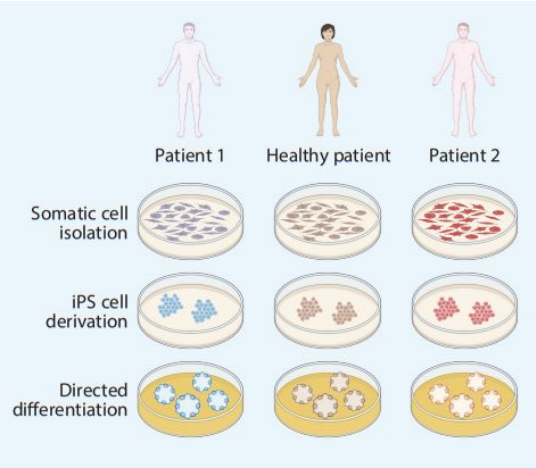
Disease Modeling

Diseases of Interest

- Acute Kidney Injury (AKI) - kidneys suddenly stop working due to damage, usually from another illness
- Chronic Kidney Disease - Progression of AKI where kidneys fail to filter the blood, excess fluid and waste build-up in the body

Considerations

- Glomerular and Tubular compartments are both affected by these diseases, matching the conditions of these cells can help to replicate a diseased kidney
    - Reduced glomerular filtration rate, thickening of compartment walls, destruction of tubule organization
    - Cells can be taken from patients to mimic these conditions more accurately



(Musah, 2024)

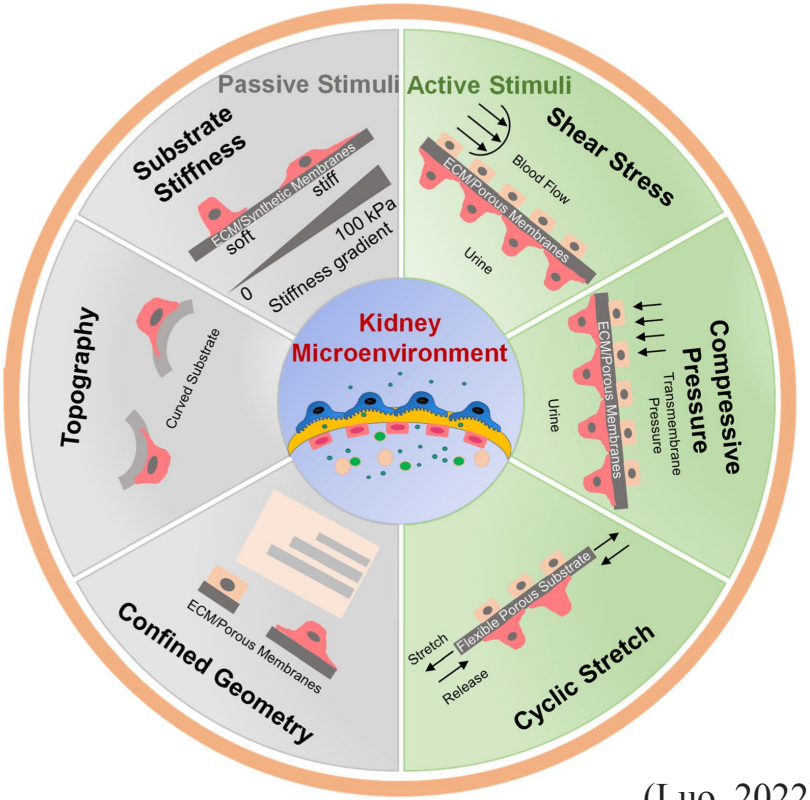
Imitation of Physiological Environment

- Healthy Kidney
  - Deliver drugs/toxins to understand how the kidney would respond
  - Use as a control for disease testing
  - Deliver substances to try and create a diseased state
- Diseased Kidney
  - Use diseased cells to create a model of AKI, chronic kidney disease, and/or renal fibrosis
  - Modeling of proteinuria
  - Testing of drugs for treatment of disease

Limitations

Advantages

- Improved physiological relevance
  - Perfusion and 3D cultures
- Multiorgan interactions are possible
- Robust and standardized fabrication techniques
  - Microfabrication
  - Microfluidics
- Microscale environment
  - Biomimetic
- Reduce need for expensive animal studies
  - Saves money and time

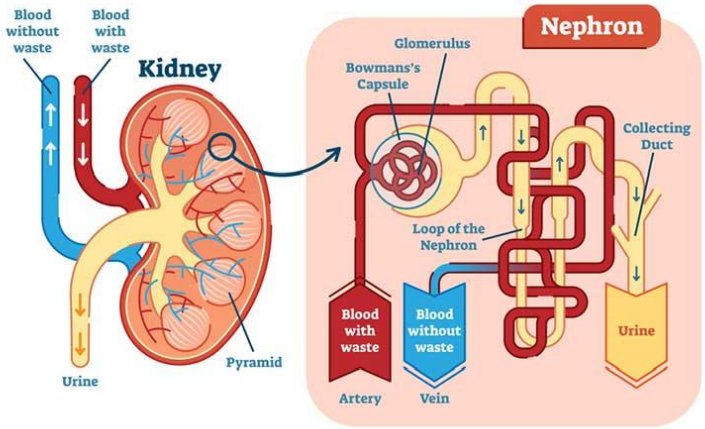


(Luo, 2022)

(MDPI, 2021)

Disadvantages

- Interactions between different renal structures is still a challenge to mimic
  - Renal tissue is very complex
- Low throughput
  - Lower volume of data that can be collected
- Lack of validation
  - Current challenge in the field
- Highly specialized
- Not compatible with some screening tools



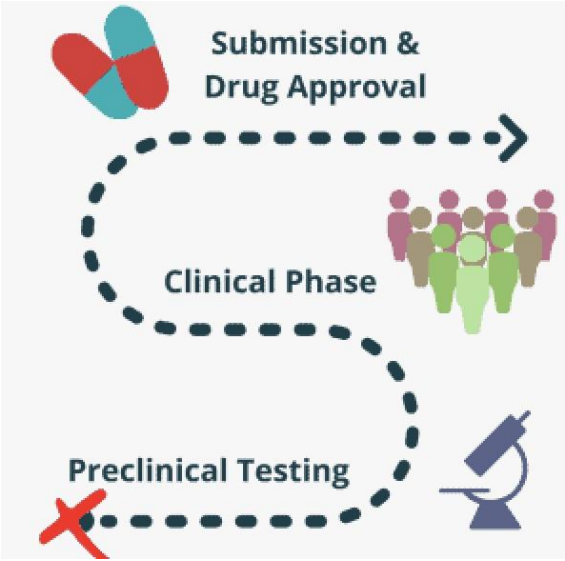
(Luo, 2022)

(Apollo Medics, 2024)

Future Directions

Drug Testing

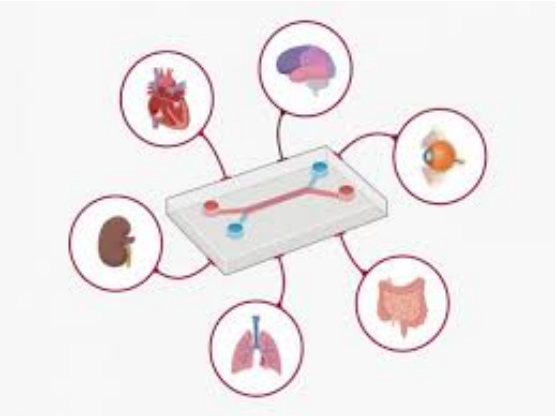
- Proximal tubule is the primary site for drug clearance
- Kidney-on-a-chip can be used to accelerate drug testing
  - Better predict drug disposition and toxicity
- New drugs must be evaluated for safety and efficacy before clinical use
  - Test the potential effects of drugs on the kidney faster than traditional methods with KoC



(Fios Genomics, 2023)

Potential Improvements

- Optimize interactions between kidney-on-a-chip and other organs to assess functionality
  - Improve tissue-tissue interactions
  - Allow us to test drugs meant for treatment of non-kidney related conditions
  - Determine if drugs are processed correctly by the kidney
- Develop more validation methods
  - How do we know the device functions as intended?



(uFluidix, 2022)

Conclusion & Summary

Conclusion

Kidney-on-a-chip has many potential applications:

- Tissue engineering
- Drug delivery
- Toxin screening

It is important to consider structural, functional, and mechanical components of the ECM to create the best biomimetic microenvironment.

There are many advantages and disadvantages of KoC technology, and challenges that have yet to be overcome.

Questions?

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