inRegen

innovation in regenerative medicine

Cell Transplant Approaches to Kidney Regeneration

AST Scientific Exchange
October 23, 2010
Orlando, FL

Relevant Financial Relationship Disclosure Statement

Sharon Presnell, Ph.D.

Tengion, Inc.

I have financial relationship(s) within the last 12 months relevant to my presentation with:

Tengion, Inc. (employee & stock holder)

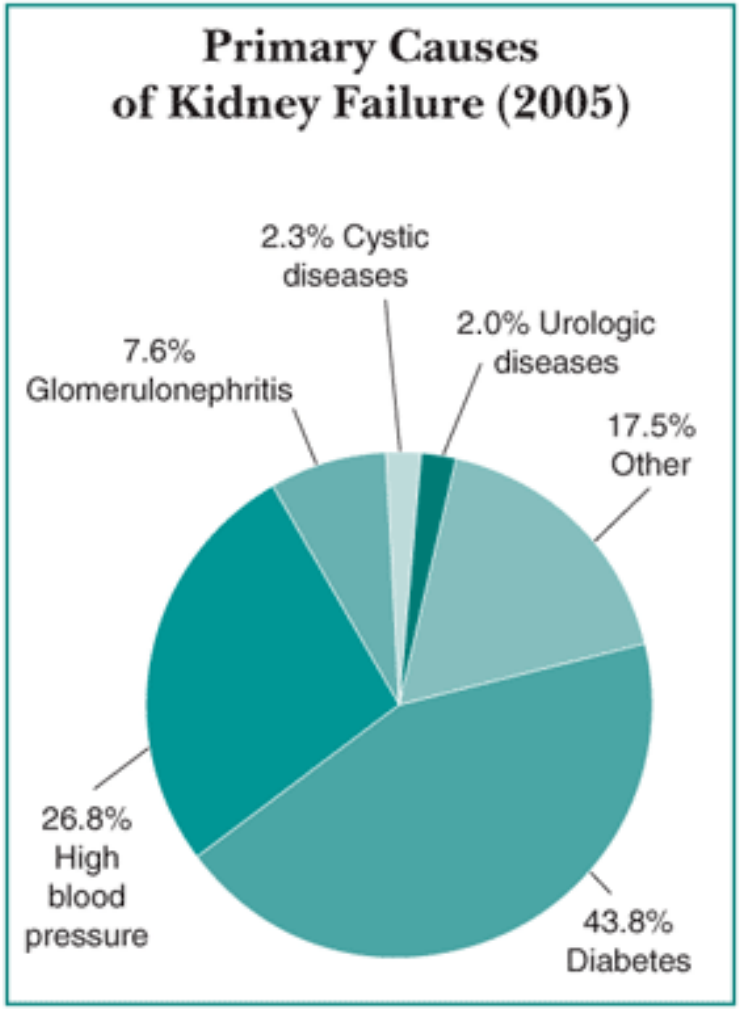
Kidney Failure can present as Acute or Chronic and can arise from many causesand can arise from many causes

Acute Renal Failure (ARF)

- *Rapidly progressive loss of renal function*
- *Oliguria (<400mL/day)*
- *Fluid imbalance*
- *Electrolyte imbalance*
- *Reversible in many patients*

Chronic Kidney Disease (CKD)

- *Develops slowly with few initial symptoms*
- *Can be consequence of unresolved acute disease (chronic after acute injury)*
- *Can be part of a disease progression (diabetes or hypertension)*
- *Staged by glomerular filtration rate (GFR)*



Treatment strategies for Acute Renal Failure

Current Standard of Care:

- ***Treat underlying cause (remove insult)***
  - *Kidney's endogenous repair mechanisms restore all / partial function*
  - *Bridging therapy (dialysis) may be required*

Emerging Technologies for ARF tested clinically:

- ***Extracorporeal device containing renal tubular cells (JASN 19:1034-1040, 2008)***
  - *Adjunct to traditional dialysis*
  - *In patients with ARF secondary to sepsis*

- ***Infusion of mesenchymal stem cells (MSC) (Nat Rev Nephrol 6:179, 2010)***
  - *Via renal artery*
  - *Ischemic injury*

Potential mechanism(s) of MSC action in Acute Renal Failure

- ***Therapeutic fusion (Held 2006 Mol Therapy 13:49)***
- ***Paracrine / cytokine-mediated modulation of inflammation and angiogenesis (reviewed by Humphreys et al., 2008 CellStemCell 2:284)***

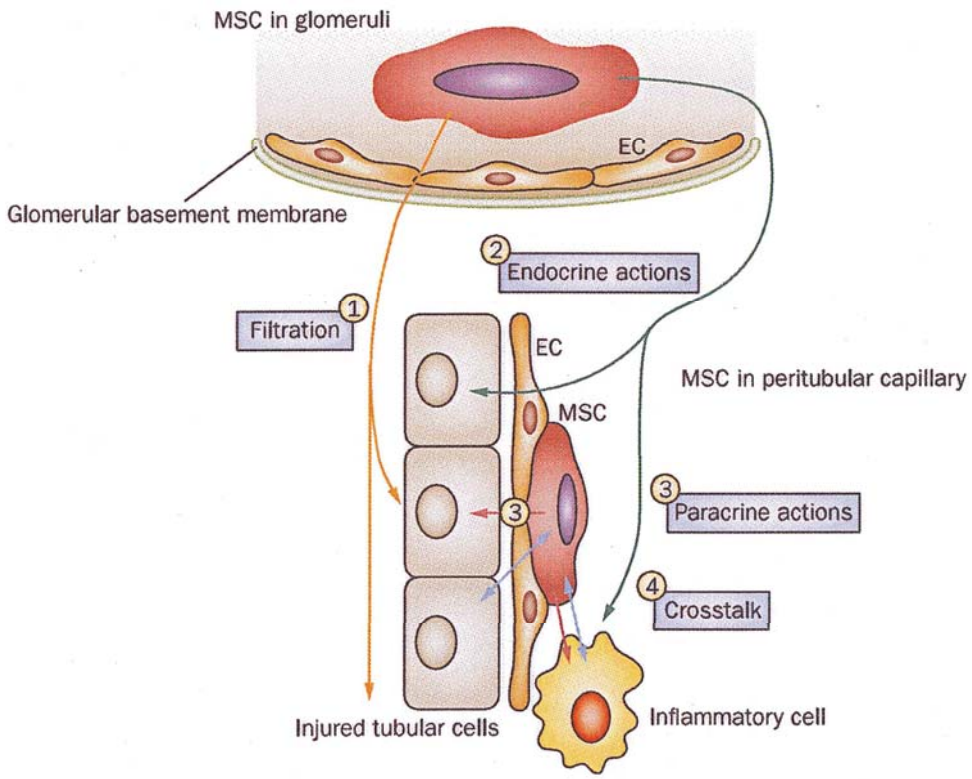
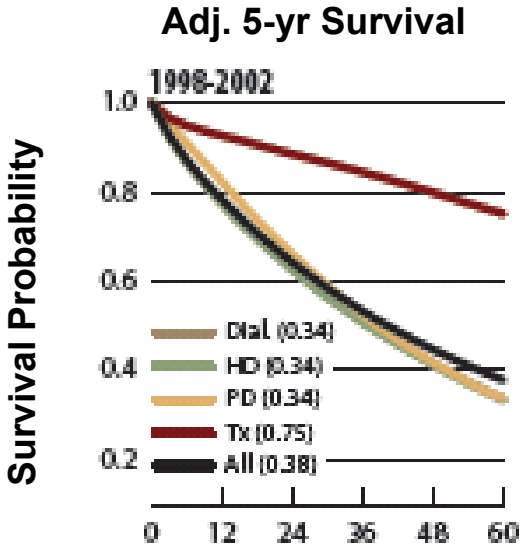
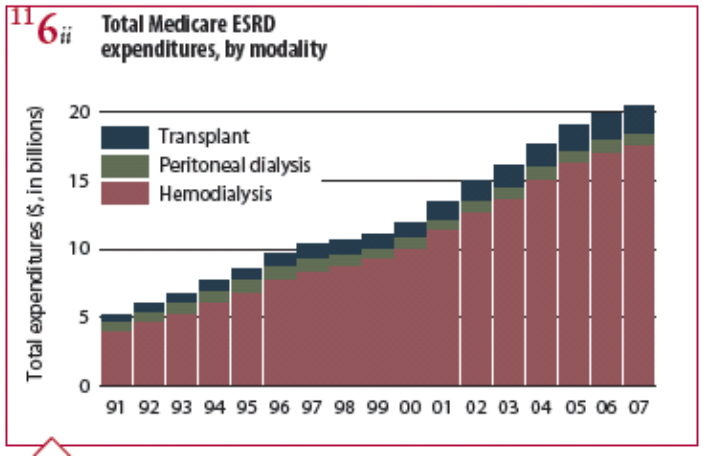


Figure from: Togel & Westenfelder C. 2010 Nat Rev Nephrol.6:179

Chronic Kidney Disease is a leading cause of death worldwide





- ***>500,000 people in the US with end-stage renal disease (ESRD)***
  - *Includes people on dialysis and those with transplants*
- ***>50,000 people with ESRD are waiting for kidney transplants in the US***
- ***>100,000 people start dialysis annually in the US***
  - *$60,000 1st year cost*
- ***>$22 billion in Medicare direct costs annually for ESRD***
  - *New treatments are needed*

Treatment strategies for Chronic Kidney Disease

Current Standard of Care:

- ***Treat underlying conditions to slow progression***
  - *Diabetes → glucose control (metformin, insulin, glitazones)*
  - *Hypertension → ACEs / ARBs*
- ***Dialysis***
- ***Transplant***

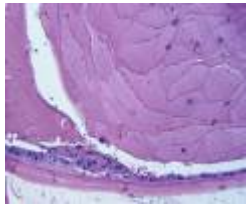
Emerging Technologies for CKD:

- ***New antifibrosis / anti-inflammatory agents undergoing preclinical & clinical testing (clinical stage)***
- ***Cell / tissue-engineering approaches***
  - *Extension of MSCs to tissue engineering approaches (Preclinical), J Nephrol. 2009 May-Jun;22(3):312-7*
  - *Use of MSCs as modulators of immune response and fibrosis (Preclinical), Stem Cells. 2009 Dec;27(12):3063-73*
  - *Autologous kidney-derived cells for regeneration of renal structure and function (Preclinical), Am J Physiol Renal Physiol. 2010 Sep 8. [Epub ahead of print]*

Tengion's products catalyze regeneration

INPUTS

Biomaterials



Cells

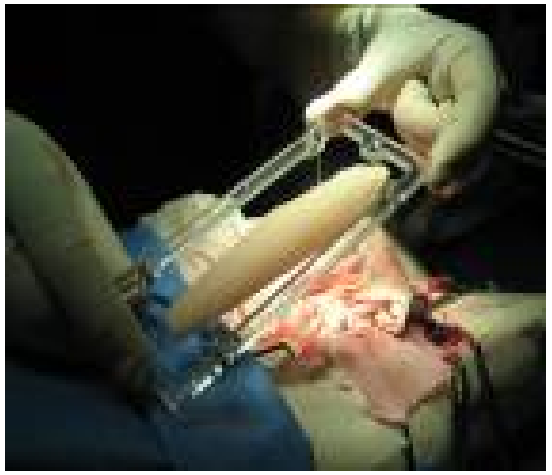
Bioprocess

Delivery Systems

INTEGRATED PLATFORM

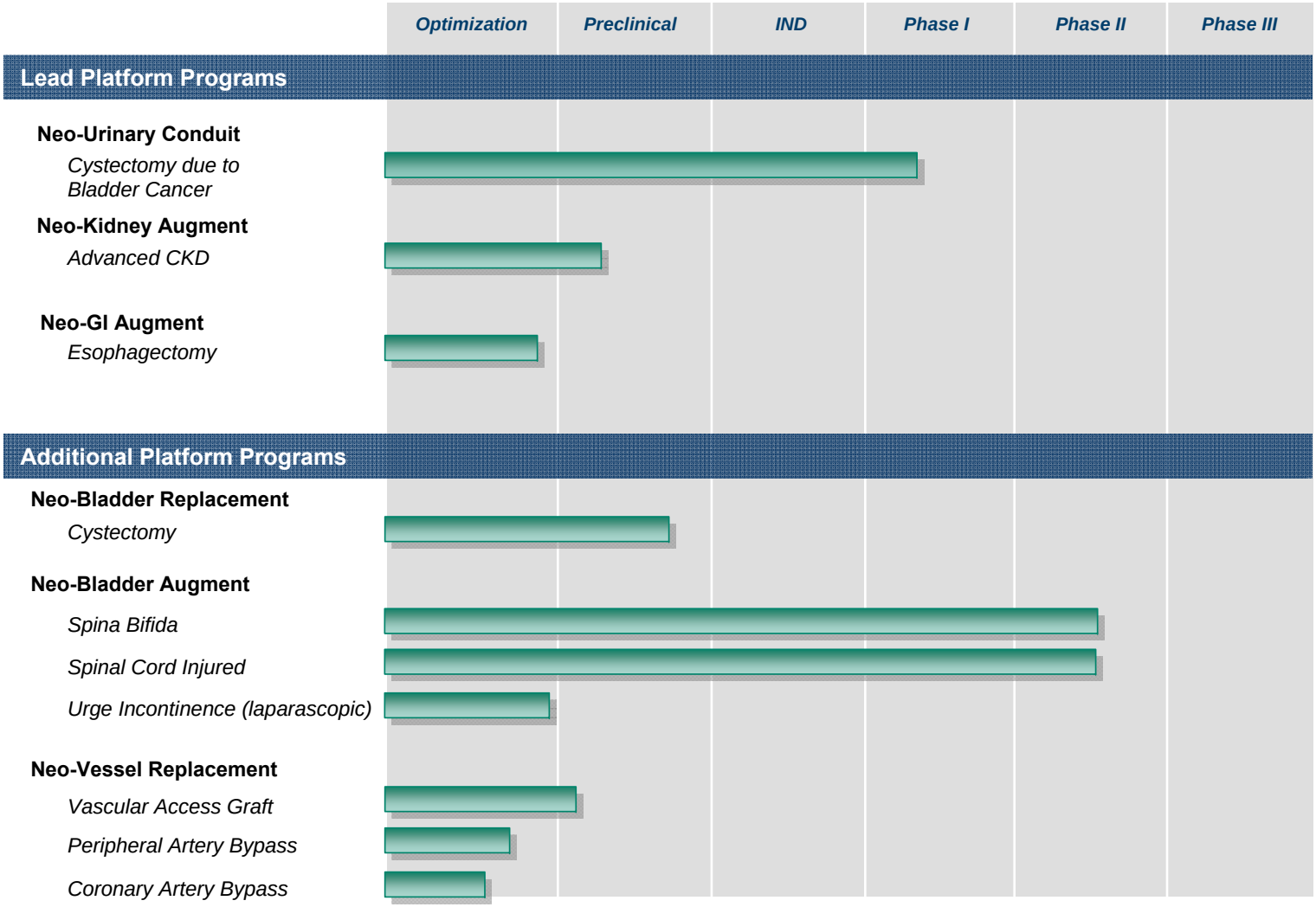
OUTPUTS

Regenerative Templates



- ***Combination products***
- ***Stimulate regeneration***
- ***Integrate into host***

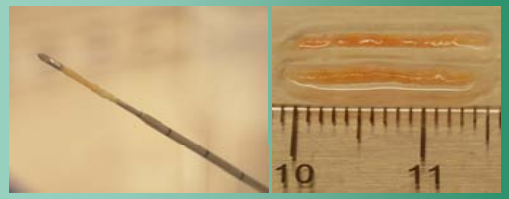
Tengion Product Pipeline



Neo-Kidney Augment™(NKA)

Applying Tengion's regenerative platform to a solid organ

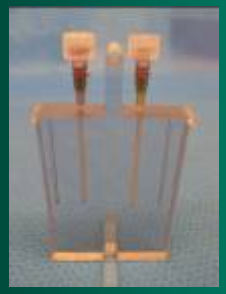
Bioactive Renal Cells from Kidney Biopsy



Isolation / Expansion of Renal Cells

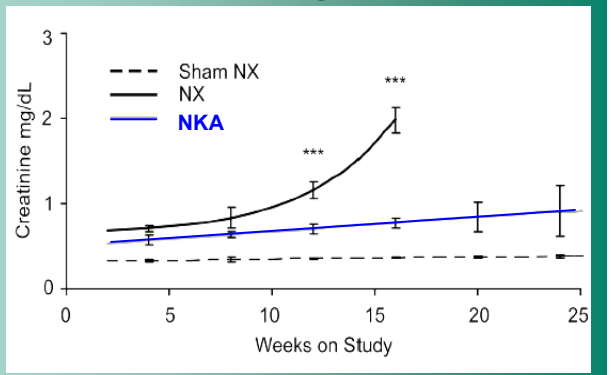
Bioreactor System for NKA Production*

Injectable Delivery System*



***In vivo* Delivery**

Functional Regeneration**

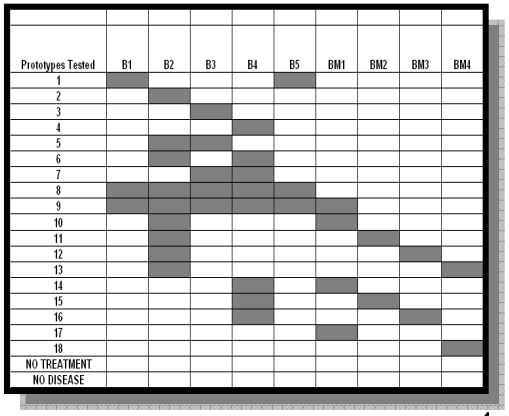


***In development**

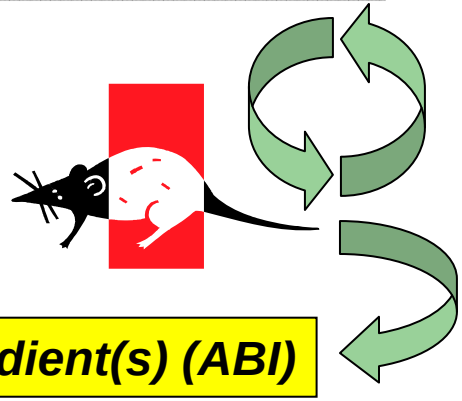
Strategic approach to identify essential components *of Neo-Kidney Augment™(NKA) prototypes*

Generate testable array of 'kidney components' based on native tissue composition

Design combinatorial experiments based on functional component characteristics



Iterative in vitro and in vivo testing in models of CKD



Identification of Active Biologic Ingredient(s) (ABI)

Validation of NKA cellular function in four models

Increases probability of success through development

1. ***NKA cells demonstrated durable function in 5/6 Nx rodent model of CKD due to renal mass insufficiency****
   - *Robust therapeutic effect (animals followed 6M post-treatment)*
   - *Reproducible across independent studies*

2. ***NKA cells function in a model of CKD secondary to obesity and Type 2 Diabetes (Active)*****
   - *Evaluation of intervention at CKD Stages 3-4 (animals followed to 1 Year of age)*
   - *NKA cells derived from diseased donors*

3. ***Will human-derived NKA cells function* in vivo? (Active)**
   - *Ischemia-Reperfusion-Gentamicin (I-R/G) model of kidney damage (chronic after acute)*
   - *Feasibility established in I-R/G model with human kidney cells*

4. ***Will the NKA cells function in a large animal model of CKD?* (Active)**
   - *Surgically-induced remnant kidney model of renal insufficiency*
   - *Feasibility established for isolation and delivery of NKA cells*
   - *Ongoing studies (active) support safety of NKA cells after intra-renal delivery*
   - *Early data support in vivo function of autologous NKA cells*

Validation of NKA cellular function in four models

Increases probability of success through development

1. ***NKA cells demonstrated function in 5/6 Nx rodent model of CKD due to renal mass insufficiency****
   - *Robust therapeutic effect*
   - *Reproducible across independent studies*

2. ***NKA cells functions in a model of CKD secondary to obesity and Type 2 Diabetes (Active)*****
   - *Evaluation of intervention at CKD Stages 3-4*
   - *NKA cells derived from diseased donors*

3. ***Will human-derived NKA cells function* in vivo? (Active)**
   - *Ischemia-Reperfusion-Gentamicin (I-R/G) model of kidney damage (chronic after acute)*
   - *Feasibility established in I-R/G model with human kidney cells*

4. ***Will the NKA cells function in a large animal model of CKD?* (Active)**
   - *Surgically-induced remnant kidney model of renal insufficiency*
   - *Feasibility established for isolation and delivery of NKA cells*
   - *Ongoing studies (active) support safety of NKA cells after intra-renal delivery*
   - *Early data support in vivo function of autologous NKA cells*

5/6 Nephrectomy model of CKD5/6

Renal disease secondary to reduced kidney mass

Rodent 5/6 Nx-induced model of renal failure

Terminal progressive renal failure

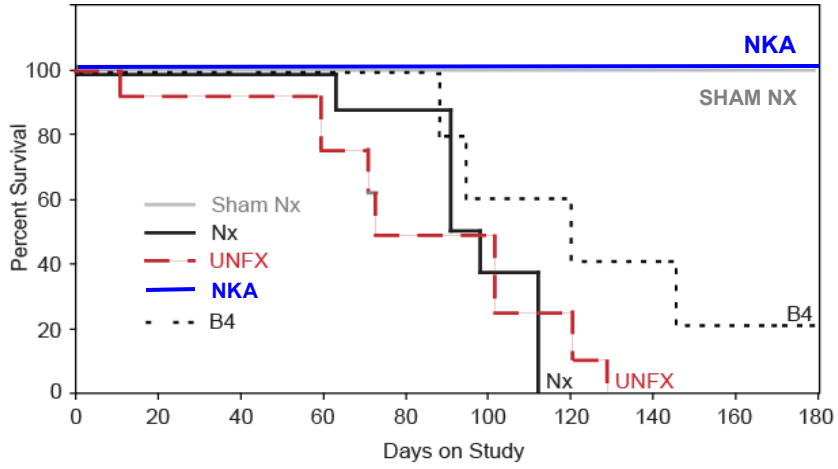
- ***0% survival at 6M post-Nephrectomy***
- ***Progressive decline in GFR***
- ***Uremic***
- ***Anemic***
- ***Proteinuric***
- ***Hypertensive***
- ***Progressive glomerular sclerosis***
- ***Progressive tubulointerstitial fibrosis***

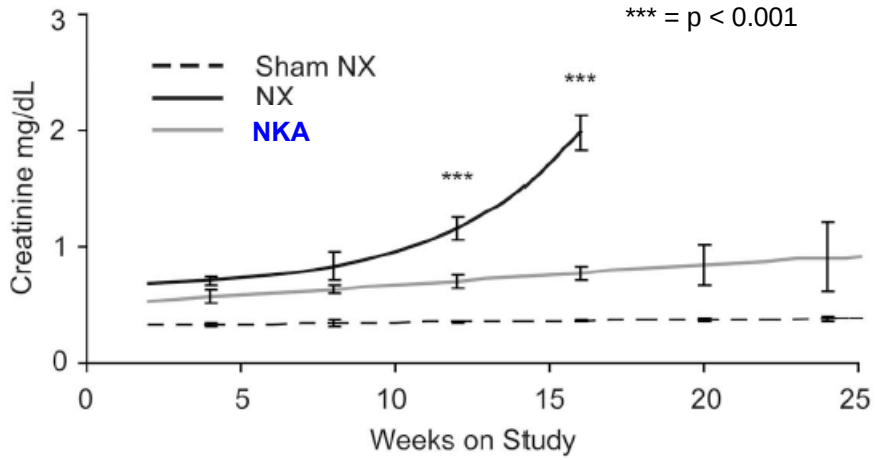
NKA cellular function validated in vivo

In rodent 5/6 nephrectomy model of chronic kidney disease

- ***NKA ABI delivered <u>after</u> chronic disease state established***
  - *sCREAT sustained at >200%*
  - *BUN sustained at >150%*

- ***Selected ABI (NKA) outperforms unfractionated mixture (UNFX) and improves multiple physiologic parameters***
  - *Enhanced Survival*
    - *(100% (NKA) vs. 0% (Nx and UNFX)*
  - *Stabilized filtration (sCreatinine)*
  - *Functional tissue regeneration*
  - *Improved protein balance*
  - *Reduced phosphatemia*





NKA provides structural and functional regeneration

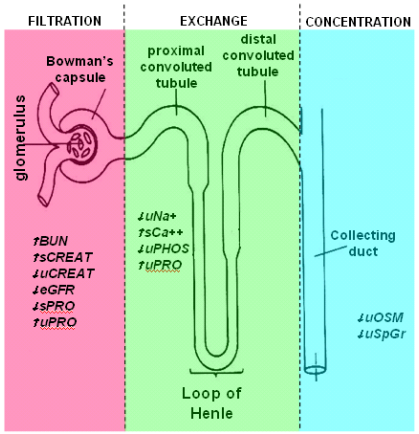
Endpoints correlate with durable tissue-level improvements

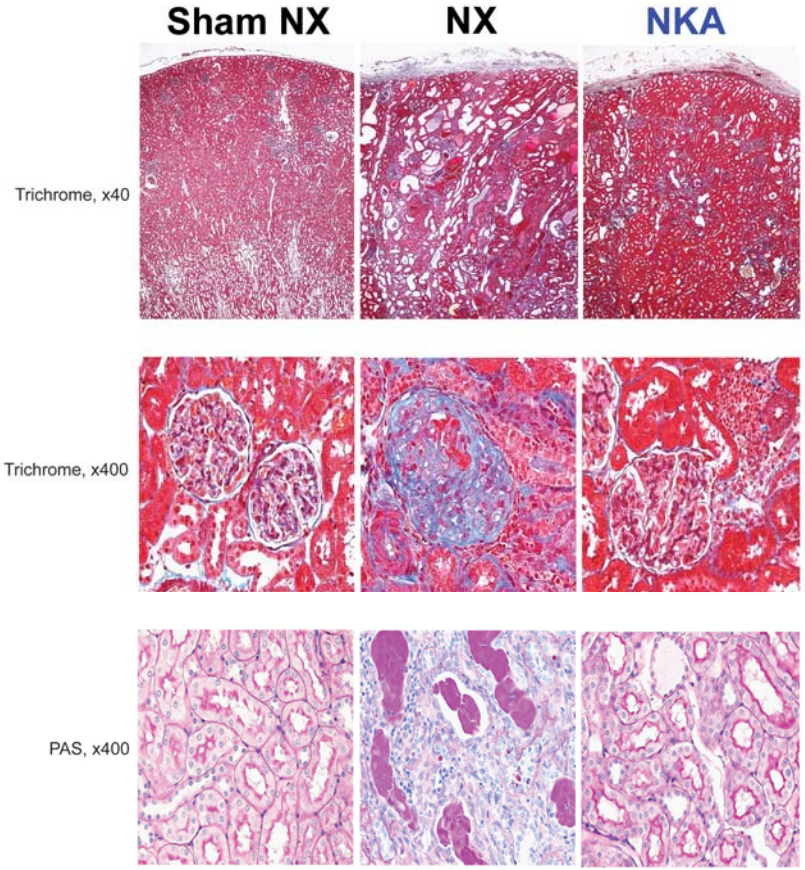
Functional regeneration

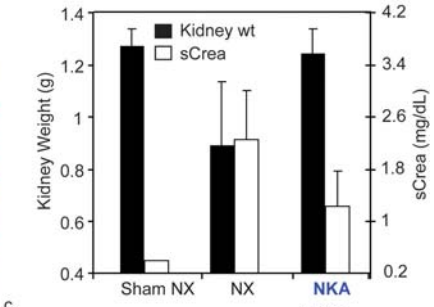
- ***Glomeruli***
- ***Tubules***

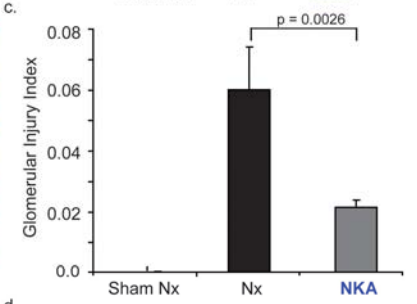
Reduced fibrosis

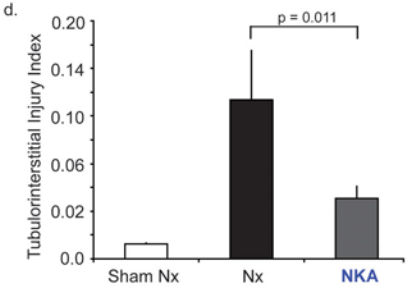
- ***Glomerular***
- ***Tubulointerstitial***











Validation of NKA cellular function in four models

Increases probability of success through development

1. ***NKA cells demonstrated function in 5/6 Nx rodent model of CKD due to renal mass insufficiency****
   - *Robust therapeutic effect*
   - *Reproducible across independent studies*

2. ***NKA cells functions in a model of CKD secondary to obesity and Type 2 Diabetes (Active)*****
   - *Evaluation of intervention at CKD Stages 3-4*
   - *NKA cells derived from diseased donors*

3. ***Will human-derived NKA cells function* in vivo? (Active)**
   - *Ischemia-Reperfusion-Gentamicin (I-R/G) model of kidney damage (chronic after acute)*
   - *Feasibility established in I-R/G model with human kidney cells*

4. ***Will the NKA cells function in a large animal model of CKD?* (Active)**
   - *Surgically-induced remnant kidney model of renal insufficiency*
   - *Feasibility established for isolation and delivery of NKA cells*
   - *Ongoing studies (active) support safety of NKA cells after intra-renal delivery*
   - *Early data support in vivo function of autologous NKA cells*

Obese ZSF1 rats model progressive nephropathy

Renal disease secondary to diabetes mellitus and hypertension

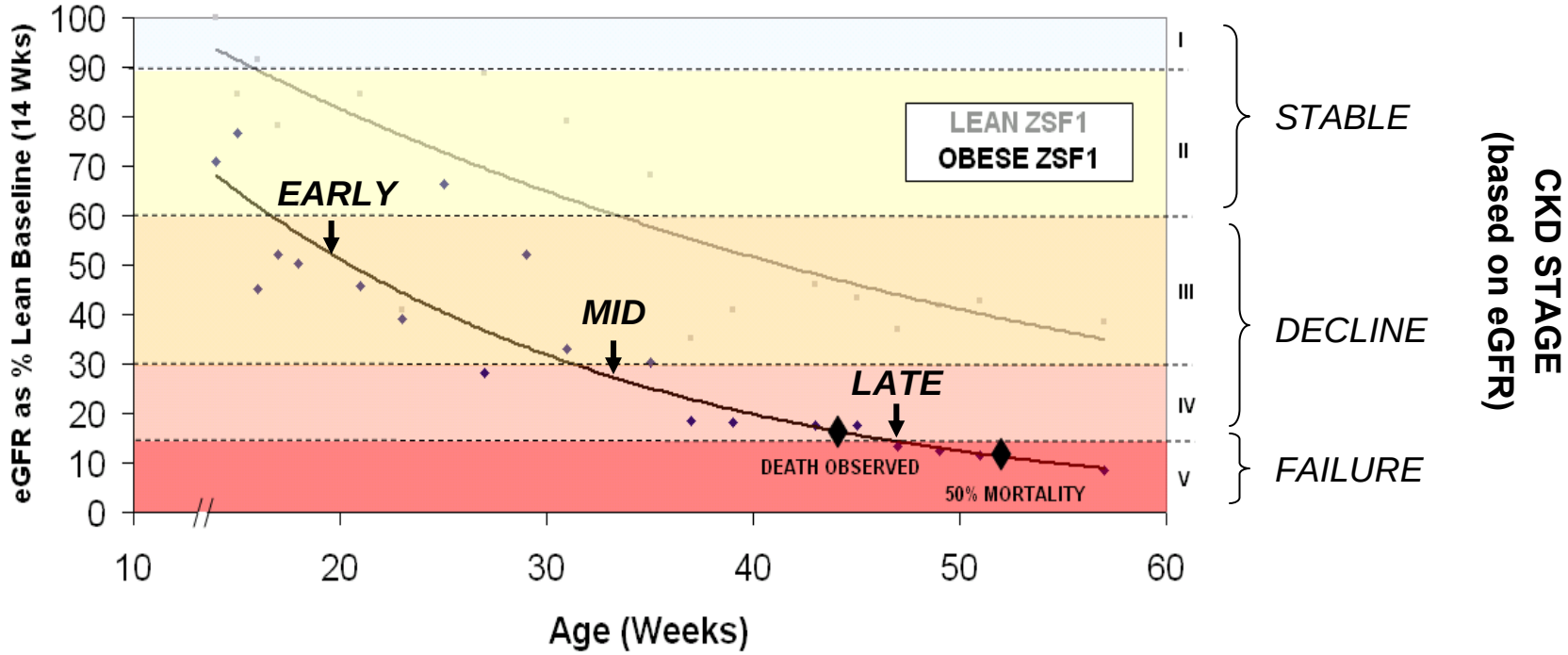
ZSF1 rodent model of Diabetic nephropathy

Aggressive Metabolic Syndrome

- ***Morbid obesity (leptin-receptor deficient)***
- ***~50% mortality at ~1yr***
- ***Multiple co-morbid conditions***
  - *Hyperglycemia*
  - *Vasculopathy*
  - *Hypertension*
- ***Progressive disease occurs throughout the nephron***
  - *Renal hypertrophy*
  - *Progressive glomerular sclerosis*
  - *Progressive decline in GFR*
  - *Tubular / interstitial fibrosis*
  - *Severe proteinuria*

Validating NKA cells in chronic disease

Renal failure secondary to obesity and Type 2 diabetes (ZSF-1)



Intervention windows:

- ***EARLY (Early Stage 3 CKD)***
- ***MID (Late Stage 3 CKD) w/ moderate control of hyperglycemia***
- ***LATE (Late Stage 4 CKD) w/ moderate control of hyperglycemia***
- ***Lean ZSF1 = positive control***

Intervention strategies:

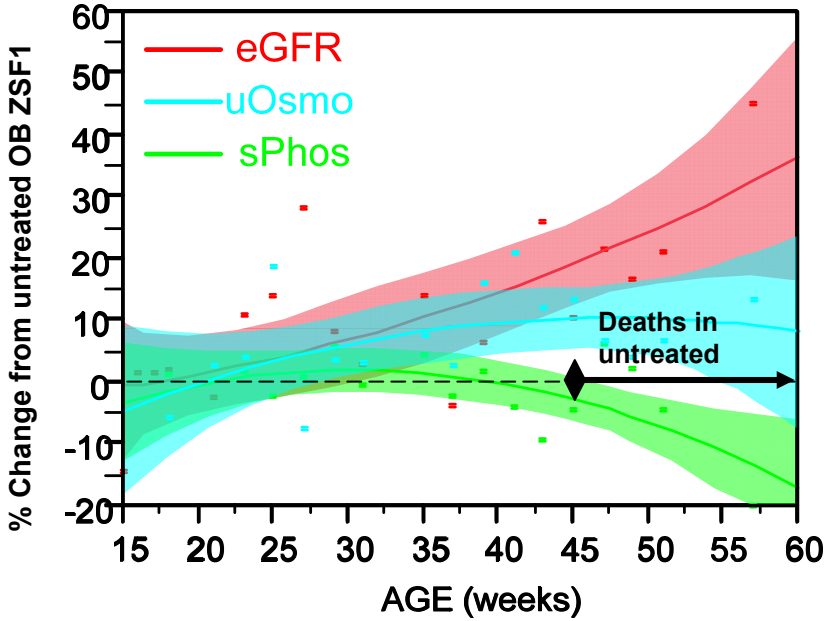
- ***Syngeneic diseased donors***
- ***Treated (1) or both kidney(s)***

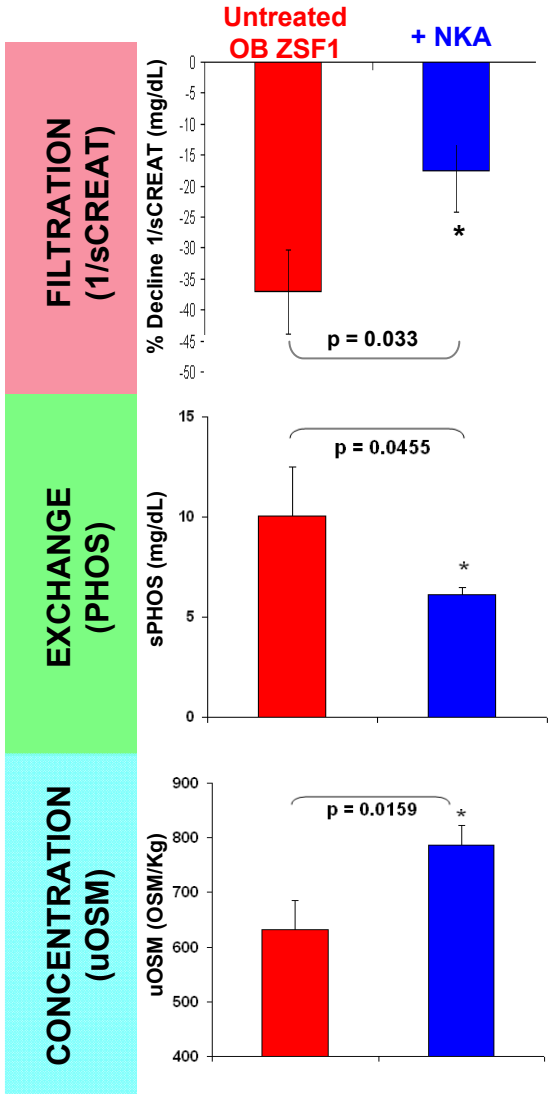
NKA cells improved function throughout the nephron

Glomeruli, tubules, and collecting ducts

At > 1 year of Age:

- **35% improvement in eGFR (Filtration)**
- **15% reduction in phosphatemia (Tubular Function)**
- **10% improvement in uOSM (Concentration)**

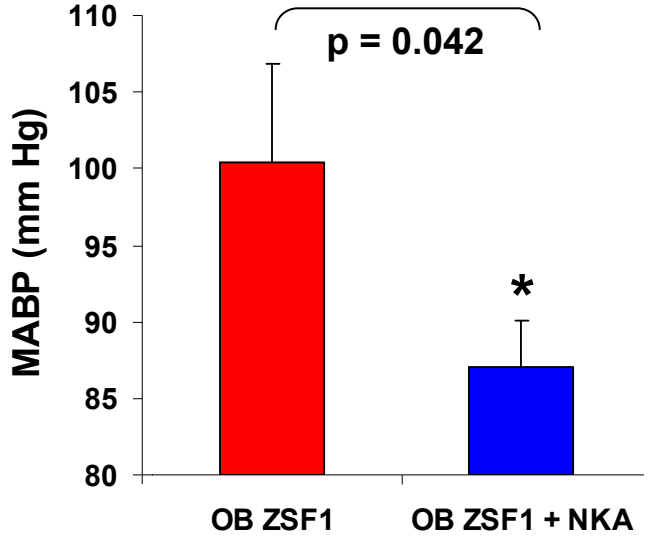




NKA cells reduced hypertension and improved survival

ZSF1 rats at >1 year of age

- ***NKA reduced mean arterial blood pressure (MABP) significantly at 57 weeks of age***



- ***NKA supports survival beyond 50% mortality time point for OB ZSF1***

Treatment Group	*63-week Survival*
OB ZSF1	20% (1/5)
OB ZSF1 + NKA ABI	100% (5/5)

Validation of NKA cellular function in four models

Increases probability of success through development

1. ***NKA cells demonstrated function in 5/6 Nx rodent model of CKD due to renal mass insufficiency****
   - *Robust therapeutic effect*
   - *Reproducible across independent studies*

2. ***NKA cells functions in a model of CKD secondary to obesity and Type 2 Diabetes (Active)*****
   - *Evaluation of intervention at CKD Stages 3-4*
   - *NKA cells derived from diseased donors*

3. ***Will human-derived NKA cells function* in vivo? (Active)**
   - *Ischemia-Reperfusion-Gentamicin (I-R/G) model of kidney damage (chronic after acute)*
   - *Feasibility established in I-R/G model with human kidney cells*

4. ***Will the NKA cells function in a large animal model of CKD?* (Active)**
   - *Surgically-induced remnant kidney model of renal insufficiency*
   - *Feasibility established for isolation and delivery of NKA cells*
   - *Ongoing studies (active) support safety of NKA cells after intra-renal delivery*
   - *Early data support in vivo function of autologous NKA cells*

Nude Rat Model of CKD

Enables functional assessment of bioactive human cells

Rodent model of CKD after acute injury

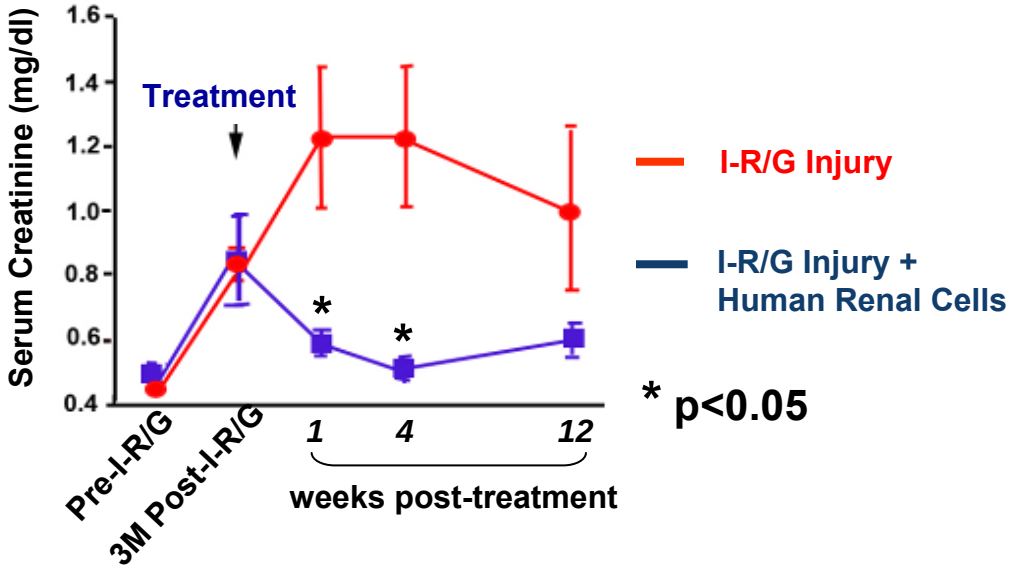
Chronic renal insufficiency

- ***Renal Failure due to Ischemia-Reperfusion + Gentamicin (I-R/G)***
- ***Rats that fail to recover 3M post-injury are utilized (Chronic after Acute)***
- ***Model characterized by stable insufficiency with partial recovery***
- ***Ischemia-Reperfusion + Gentamicin injury in nude rats enables functional testing of human cellular components***

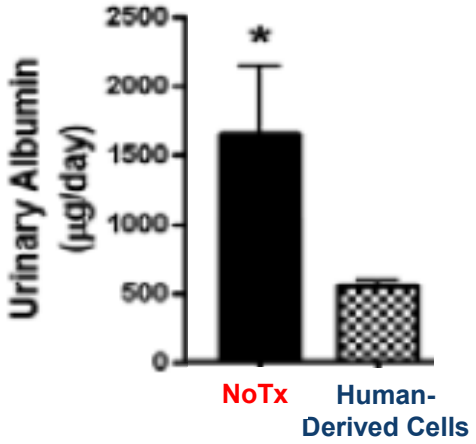
Human Kidney Tissue Regeneration in nude rats

Delayed progression of CKD and stabilized renal function

Human-derived cells prevent renal failure in CKD Nude Rats for 3 mo.



Human-derived cells improve CKD Nude Rat nephron function



Validation of NKA cellular function in four models

Increases probability of success through development

1. ***NKA cells demonstrated function in 5/6 Nx rodent model of CKD due to renal mass insufficiency****
   - *Robust therapeutic effect*
   - *Reproducible across independent studies*

2. ***NKA cells functions in a model of CKD secondary to obesity and Type 2 Diabetes (Active)*****
   - *Evaluation of intervention at CKD Stages 3-4*
   - *NKA cells derived from diseased donors*

3. ***Will human-derived NKA cells function* in vivo? (Active)**
   - *Ischemia-Reperfusion-Gentamicin (I-R/G) model of kidney damage (chronic after acute)*
   - *Feasibility established in I-R/G model with human kidney cells*

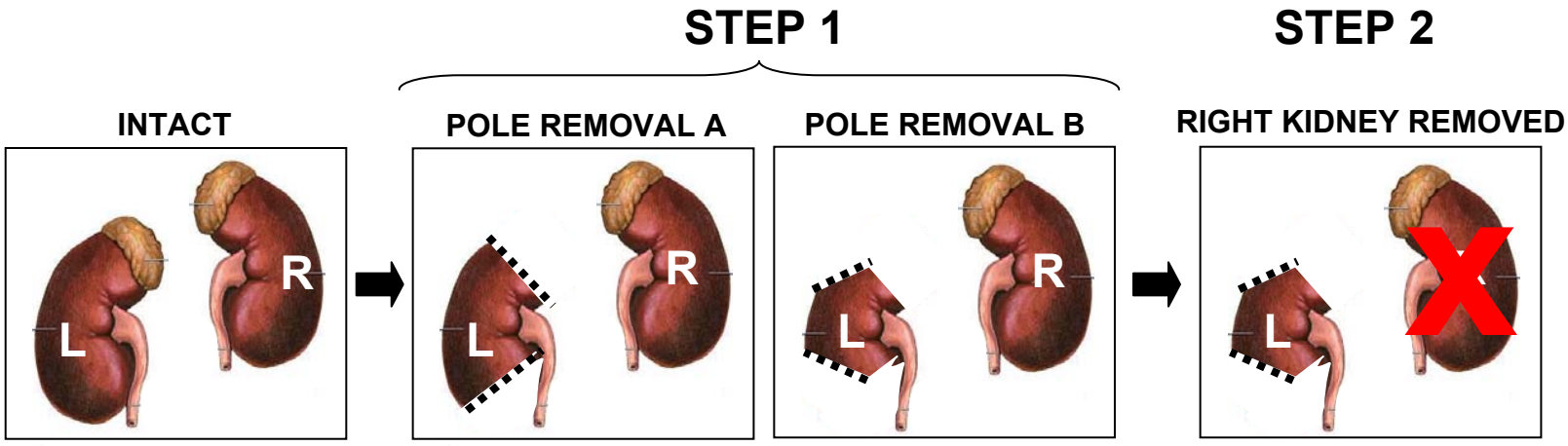
4. ***Will the NKA cells function in a large animal model of CKD?* (Active)**
   - *Surgically-induced remnant kidney model of renal insufficiency*
   - *Feasibility established for isolation and delivery of NKA cells*
   - *Ongoing studies (active) support safety of NKA cells after intra-renal delivery*
   - *Early data support in vivo function of autologous NKA cells*

Modified 5/6 Nephrectomy model of CKD

Renal disease secondary to reduced kidney mass

Canine 5/6 Nx-induced model of renal failure

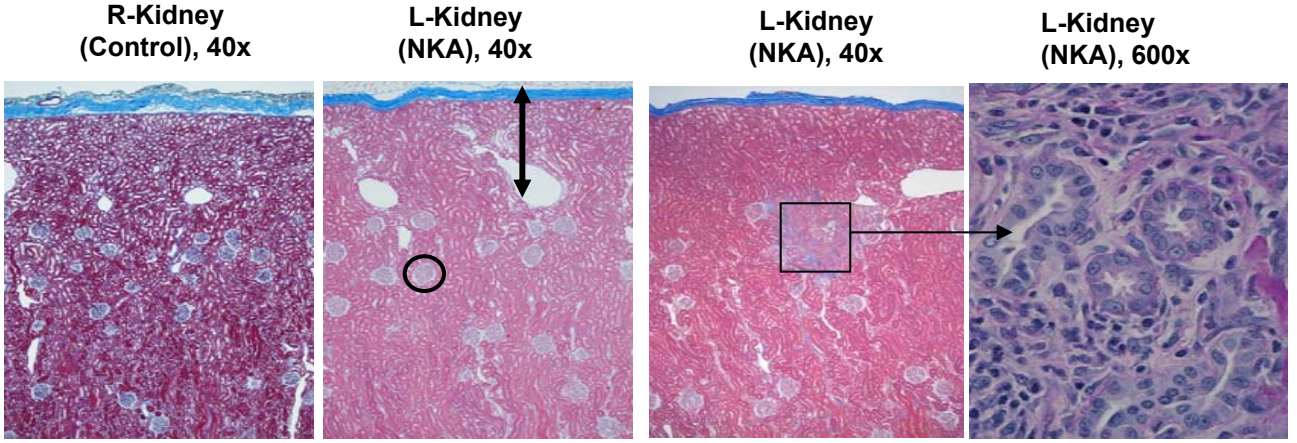
- **Chronic renal insufficiency**
  - ***>50% reduction in GFR***
  - ***Mild hypertension***
  - ***Uremia***
  - ***Mild anemia***
  - ***Progressive proteinuria***
  - ***Gradual weight loss***



Safety of NKA delivery verified

In dogs with single kidney (n=2)

- ***No adverse events***
- ***No systemic evidence of renal injury***
  - *serum chemistry*
- ***Normal histology (1M)***
  - *Normal post-nephrectomy compensation*
  - *Foci of tubular regeneration at delivery site*

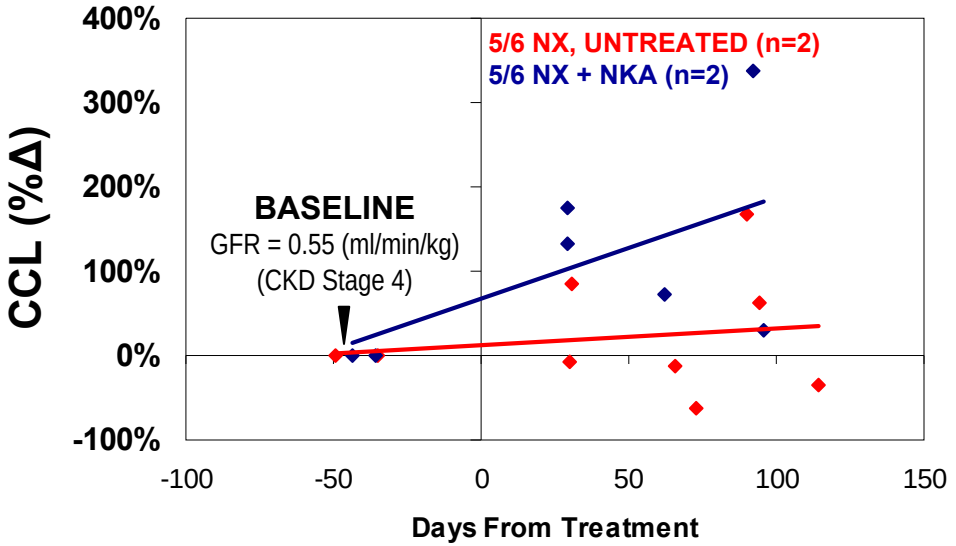


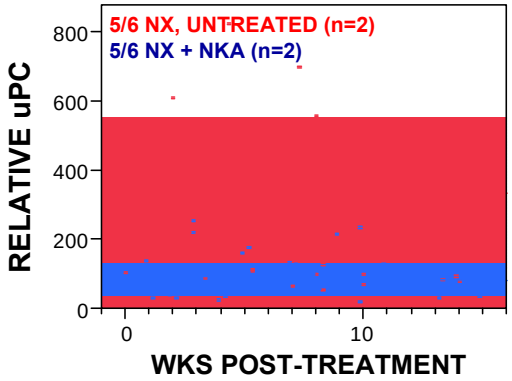
Pilot study indicates Renal Cells improve GFR

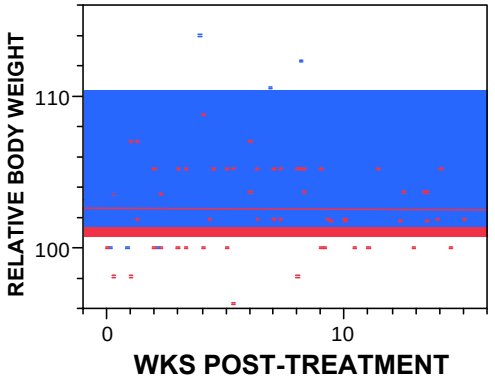
In 5/6-Nephrectomized dog model (ACTIVE)

Treatment with NKA shows trends of efficacy:

- **Increased Creatinine Clearance (CCL)**
- **Reduced Urine Protein:Creatinine Ratio (uPC)**
- **Increased Body Weight**



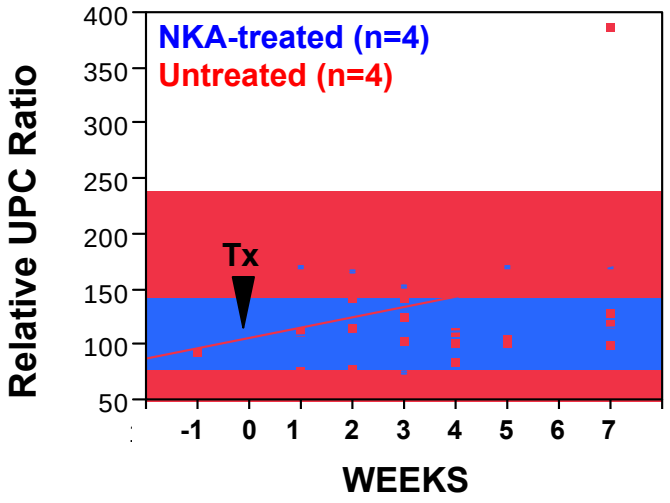




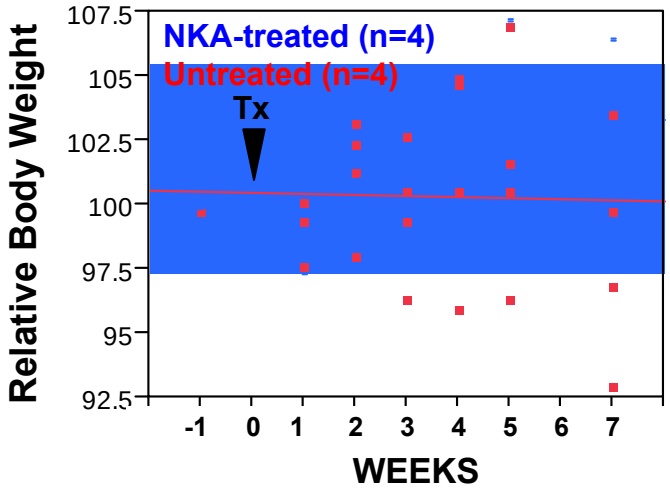
Controlled follow-up study confirms NKA function

Significant improvement within 7 weeks of treatment

Renal filtration & protein balance improve with treatment (p = 0.0281)*



Treatment promotes weight gain vs. weight loss (p < 0.0001)*

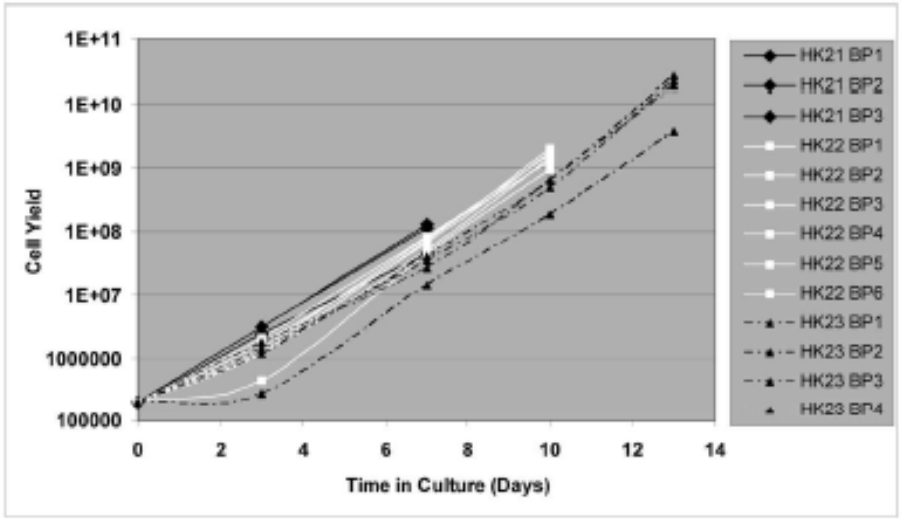


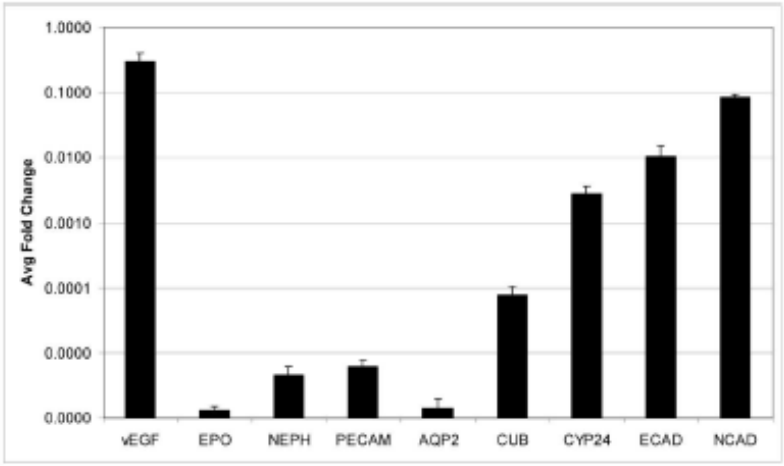
*Multivariable linear regression using 'Treatment', 'Age', and 'Animal' to predict UPC and Body Weight. P values = Effects Test for Treatment.

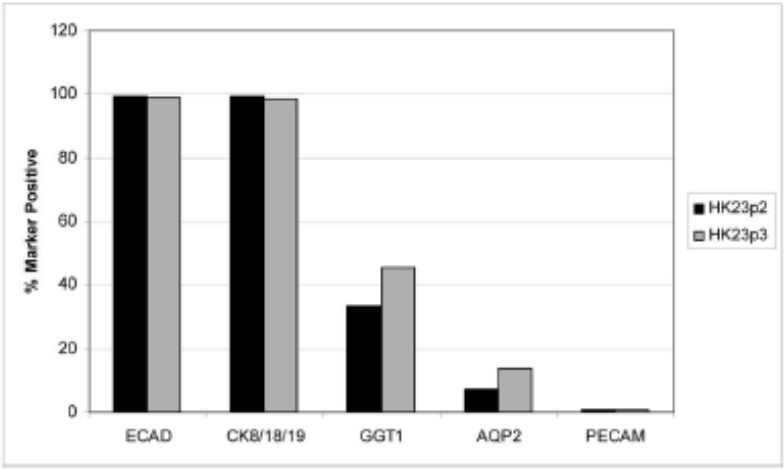
Human translation of NKA cellular components

Isolation, characterization, & expansion from human CKD-derived kidney tissue

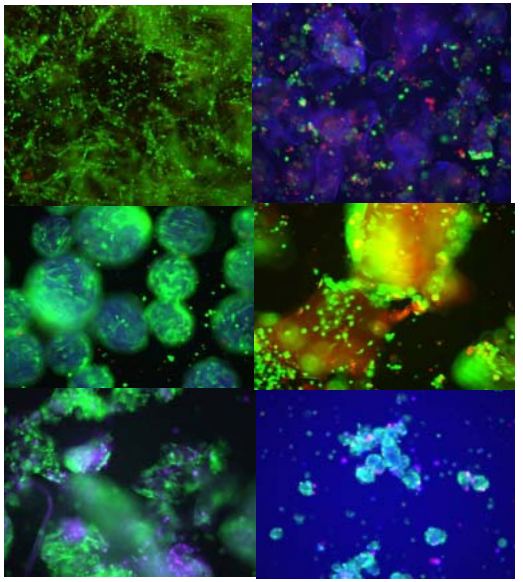
- *Standard core needle biopsy procedure (0.02g tissue)*
- *Cells can be expanded and cryopreserved*
- *Salient phenotypic attributes are preserved*
- *Supports autologous sourcing strategy*







Development of NKA product candidate
To target delivery, increase stability, and expand use

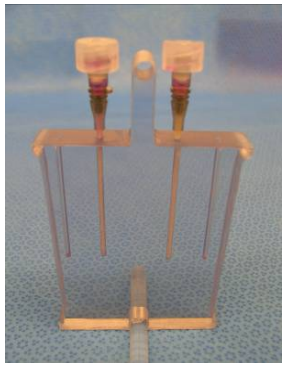


Product Candidate

- Formulated for targeted delivery, stability, durability and function of the ABI

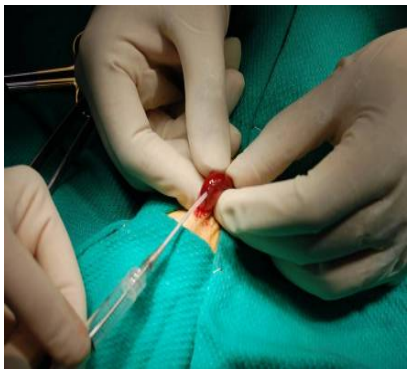
Delivery System

- Bioreactor system for stable transport and delivery of product



Delivery Procedures

- Laparotomy (open procedure)
- Laparoscopic (ultrasound guided)



SUMMARY / NKA PRECLINICAL RESULTS

- ***Proof-of-concept established that NKA improves renal function compared to untreated controls in (3) rodent models of CKD***
  - *5/6 Nephrectomy model of terminal renal insufficiency (6M)*
  - *ZSF1 model of metabolic syndrome and diabetic nephropathy (1 YR)*
  - *Human cells in a nude rat model of I-R/G, chronic after acute renal failure*

- ***Early observations from ongoing large animal studies are consistent with early small animal results***
  - *Isolation and delivery of NKA cells from canines with renal insufficiency*
  - *Early results indicate in vivo function of NKA cellular components*

- ***Isolation of NKA ABI established from human kidneys with CKD***
  - *Reproducible isolation from core needle biopsies*
  - *Supports autologous sourcing strategy from target patient population*

CONCLUSIONS

- ***Preclinical / clinical advancements have been made in the development of cells / pharmaceuticals that facilitate endogenous regenerative processes in Acute Renal Failure***

- ***Regenerative approaches to the treatment of Chronic Renal Failure present significant challenges***
  - *Considerations of degenerating tissue architecture*
  - *Significant co-morbid conditions in patient populations*

- ***Significant preclinical progress is being made in the development of cell-based approaches to the treatment of CKD***
  - *Cells as modulators of inflammation and fibrosis*
  - *Cell-based products that stimulate regeneration*


inRegen
innovation in regenerative medicine