From Pathogenesis to Device Development: How can we best use animal models?

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Outline
- Why interventional nephrologists need animal models
- Describe standard large and small animal models of AVF and AVG stenosis
  - Elucidate underlying mechanisms of stenosis
  - Develop novel devices to reduce dialysis access stenosis
- Why animal models need interventional nephrologists

Why do we need animal models in interventional nephrology
- Dialysis access stenosis is a unique situation!!!
- No other specialty deals creates an “iatrogenic” connection between an artery and vein!!
- Focus on venous rather than arterial stenosis
- Uremia is a complicating factor
- Complete lack of effective therapies (need to elucidate mechanisms and to test out novel devices)

A vein is not an artery
- Anatomical: Veins have a poorly defined internal elastic lamina
- Physiological: Veins release less nitric oxide and prostacyclin
- Molecular: Significant differential expression of gene products between normal vein and artery

Animal models of AVF and AVG stenosis

LARGE
- Pig AVF/AVG
- Goat AVF/AVG
- Sheep AVG
- Dog AVG

SMALL
- Mouse AVF
- Rat AVF

Size Matters!!

LARGE
- Surgical techniques
- Devices
- Grafts
- Bioengineered vessels
- Surgical configurations

SMALL
- Mechanisms
- Knock out or transgenic animals
- Easy availability of antibodies and gene sequences
- COST!!!!
Development of a pig model of venous neointimal hyperplasia in PTFE dialysis access grafts

Suitable vascular tree
Response of vessel wall to injury is similar to Man

Specimen Preparation

Validation Time Points: Days 2, 4, 7, 14, 28

Pig Model (Day 2)

Pig Model (Day 14)

Significant venous neointimal hyperplasia at 28 days

Human/Pig Similarities

Human GVA  Pig GVA
External radiation attenuates neointimal hyperplasia

- Single dose of 16 Gy
- 5 x 6 cm area
- Within 24 hrs of surgery

Vein: 50.5% vs 38.4%
p = 0.021
24% reduction

Placement of Perivascular Paclitaxel Polymers into a Pig Model of Venous Stenosis

The pig is ideally suited for devices (Sutureless anastomosis graft)

Sutureless Anastomosis Hybrid Graft

Optiflow™ is a sutureless anastomotic conduit for AVF creation

Optiflow anastomotic conduit
Optiflow Angiography

AV Fistula Model Development (Dissection)

AV Fistula Model Development (Arteriotomy)

AV Fistula Model Development (Anastomosis)

AV Fistula Model Development

Neointimal budding in the 7d AVF

Wang et al. NDT 2008

Krishnamoorthy et al. P-PO1054, ASN 2006
Aggressive stenosis at 42d

Alternative Origins for Neointimal Cells: Role of the Adventitia

Eccentric macrophage infiltration at 2d

Eccentric neointimal hyperplasia at 42d

AV Fistula Model Development (Shear Stress Analysis)

Eccentric shear stress to macrophages to stenosis

Oscillatory shear stress

Inflammatory (macrophage) response

Chemokines and cytokines

Intima-media thickening
The Pig is a good large animal model

- Response to injury similar to humans
- Animal model of choice for cardiovascular research (comparisons)
- Better availability of antibodies and gene sequences as compared to dogs and sheep
- More aggressive response to injury is an advantage in intervention studies
- Lack of thrombosis could be a disadvantage (Dog)

Can we make a uremic pig??

- Yes
- Misra et al. use embolization of one kidney followed by selective renal artery infarction
- We have not made pigs uremic
- Remember you don’t want them uremic enough to require HD

Mouse Model of AVF stenosis (14d)

Mouse Model Creation

Uremic mouse model of AVF stenosis

- Cauterize right kidney
- Nephrectomy left kidney 2 wks later
- Wait 6 weeks (BUN = 50-70)
- Place AVF

Avoid morbidity and mortality due to uremia

Uremic mice have increased AV fistula stenosis

- Choi et al. JASN. 2008
The Mouse is a good small animal model

- Knock outs
- Transgenics
- Gene sequences
- Molecular tools

Surgery is NOT easy
A good human surgeon is not always a good mouse surgeon!!

Do we really need animal models

A Mouse is not a Man and neither is a Pig!

Do we need animal models?

- Yes for safety
- Yes for technique and feasibility
- Yes for mechanisms
- Yes for an efficacy signal

Do we need animal models?

- Proof of final human efficacy can only come from human clinical trials!!

Animal models and interventional nephrology: a win-win situation

- Animal models need intervention skills
- New devices will mean new angiographic appearances
- Animal model work gives you a heads up

Animal models and interventional nephrology: a win-win situation

- Increases innovation
- Develop and test out new devices
- Intellectual property
- Starting a company!
- Go from an idea to a product!!
Paclitaxel Polymers Inhibit Neointimal Hyperplasia

Kelly et al., Nephrol Dial Transplant 2006

Periadventitial injection

- Endovascular device such as the “Bullfrog” microinfusion catheter (Mercator-Med)

Tailor therapies to the biological course of vascular stenosis

Drug A initially followed by Drug B at 6 monthly intervals

AXZ-100

Bullfrog Catheter Injections
PTFE grafts coated with Cu-II coatings generate a significant NO flux POST-IMPLANT.

Is there a difference in graft thrombosis?

Hemodynamics and Histology

AV Fistula Model (CT Angiography)

3D Mesh Framework
AV Fistula Model (Flow and Pressure Analysis)

Flow
Pressure

3D Wall Shear Stress Profile

Proximal Artery
AV Anastomosis
Proximal Vein

Wall Shear Stress Alterations in Systole and Diastole

Implant Design

Conduit (Vein)
Flange (Artery)
"Heel Flange"
"Toe Flange"
"Side Flange"
Do we really need animal models

A Mouse Is not A Man!

Do we need animal models

• Yes for safety
• Yes for technique and feasibility
• ? for efficacy

AV Fistula Model Development (Curved)

Optiflow Angiography

Optiflow™ is a sutureless anastomotic conduit for AVF creation
1. Mobilize vessels

2. Venotomy

3. Insert device into vein

4. Suture the vein and mark arteriotomy

5. Arteriotomy

6. Insert flanges inside arteriotomy

The adventitia is important!

Mouse model of AVF stenosis

- End of jugular vein to side of carotid artery
- 11/0 suture; finer than a human hair; barely visible
- Significant neointimal hyperplasia by 2 wks
- Some thrombus formation also